Recommendation and Guideline

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Diagnosing Hepatocellular Carcinoma Using Sonazoid Contrast-Enhanced Ultrasonography: 2023 Guidelines From the Korean Society of Radiology and the Korean Society of Abdominal Radiology

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Sonazoid, a second-generation ultrasound contrast agent, was introduced for the diagnosis of hepatic nodules. To clarify the issues with Sonazoid contrast-enhanced ultrasonography for the diagnosis of hepatocellular carcinoma (HCC), the Korean Society of Radiology and Korean Society of Abdominal Radiology collaborated on the guidelines. The guidelines are de novo, evidence-based, and selected using an electronic voting system for consensus. These include imaging protocols, diagnostic criteria for HCC, diagnostic value for lesions that are inconclusive on other imaging results, differentiation from non-HCC malignancies, surveillance of HCC, and treatment response after locoregional and systemic treatment for HCC.
Keywords: Guideline; Ultrasound, Contrast-enhanced; Hepatocellular carcinoma; Sonazoid; Perfluorobutane

INTRODUCTION

Sonazoid® (perfluorobutane; GE Healthcare) is a second-generation ultrasound contrast agent approved for clinical use in China, Japan, Korea, Norway, Singapore, and Taiwan. The approved indication for Sonazoid in these countries is contrast-enhanced ultrasonography (CEUS), which is used to characterize focal hepatic disease in adults [1].

Although all second-generation agents have similarities, Sonazoid is a combination of blood-pool and Kupffer cell contrast agents, unlike pure blood-pool contrast agents. The Asian Federation of Societies for Ultrasound in Medicine and Biology published consensus statements and recommendations for the clinical use of Sonazoid, based on expert opinions and several consensus meetings [2]. These publications addressed the general characteristics of Sonazoid and the typical imaging appearance of common focal liver lesions. However, they did not include diagnostic

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criteria or performance evaluations for Sonazoid CEUS, and it remains unclear whether the Liver Imaging Reporting and Data System (LI-RADS) can be applied. Although a recent meta-analysis on the diagnostic performance of Sonazoid [3] reported a pooled sensitivity and specificity of 90% and 97%, respectively, the included studies did not use the same diagnostic criteria for hepatocellular carcinoma (HCC), on which the diagnostic performance for HCC depends.

In other words, two fundamental questions regarding the diagnostic criteria for HCC using Sonazoid CEUS remain unanswered. The first question concerns the applicability of the major imaging features of HCC, such as specific arterial phase hyperenhancement (APHE) and washout, to Sonazoid CEUS. The second concern is the role of Kupffer phase imaging in the diagnosis of HCC. Because most Sonazoid microbubbles are taken up by Kupffer cells in the hepatic sinusoid, Sonazoid CEUS should be considered to show greater enhancement of the background parenchyma in the delayed phase than in other CEUS studies. To conduct reliable research on Sonazoid, the terminology should be standardized, and guidelines for CEUS studies of patients at risk of HCC should be established.

In 2021, with support from the Korean Society of Radiology and Korean Society of Abdominal Radiology (KSAR), 20 Korean abdominal radiologists with expertise in CEUS developed guidelines for diagnosing HCC using Sonazoid CEUS based on eight key questions. Guideline development took approximately 1 year (May 2021 to May 2022), and the guidelines were presented at the KSAR Annual Meeting. They were also shared with related associations such as Korean Liver Cancer Association (KLCA) and Korean Society of Ultrasound in Medicine (KSUM). The goal of these guidelines was to provide an evidence-based standard for the diagnosis of HCC using Sonazoid CEUS.

Guideline Development Methodology

Literature Search

Given the limited number of published clinical studies on Sonazoid, all pertinent articles discovered were gathered as potential evidence for guideline development. Articles relevant to each of the key questions were selected and analyzed by their respective development groups.

A systematic literature search of publications in English was performed by an expert radiologist (S.H.C.) and an experienced research librarian. The databases used for searching were MEDLINE, Embase, and the Cochrane Library. The search keywords used were HCC, ultrasound, and Sonazoid (or Kupffer phase). The search was conducted on October 15, 2021. The exclusion criteria were as follows: 1) it was not written in English, 2) it was not an original research or a systematic review or meta-analysis, 3) the full text of the article could not be obtained, and 4) it was not a human study. In that search, 573 studies were retrieved from the databases, of which 403 remained after duplicate removal. After the first eligibility evaluation was performed by the researchers (H.J.K., S.L., Y.S.C., and J.A.H.), 241 studies were further analyzed (Fig. 1).

Developing Key Questions

Seven teams (developing groups) developed the key questions. Group 1 made recommendations for two closely related questions regarding the diagnostic criteria for Sonazoid CEUS. Each of the remaining groups provided recommendations. Each team searched for relevant studies in the literature pool (n = 241) and designated them as evidence tables (Supplementary Tables 1–7). In this process, 45 studies were selected by the developing groups. All selected studies were evaluated by a radiologist experienced in guideline development (W.K.J.) using the Quality Assessment of Diagnostic Accuracy Studies-II (Supplementary Tables 1–7, Supplementary Fig. 1).

Recommendation Statements

The recommendation statements and their levels were designed by the developing groups based on the selected evidence. The evidence levels of the statements were based on those of the Oxford Centre for Evidence-based Medicine (Table 1) [4].

Consensus

To ensure consensus, two consensus meetings were held (April 21, 2022, and May 11, 2022) by teleconference (Zoom, Zoom Video Communications). A web voting system (Naver Office) was used for the blinded voting. All recommendation statements were discussed and approved by all developing members using the Delphi method. Six grades of agreement were used: 1) strongly disagree, 2) disagree with major reservation, 3) disagree with minor reservation, 4) agree with major reservation, 5) agree with minor reservation, and 6) strongly agree. If more than 80% of the participants scored a statement as either agreeing with minor reservation or strongly agreeing (5 or 6), it was considered a consensus, and the recommendation was accepted.

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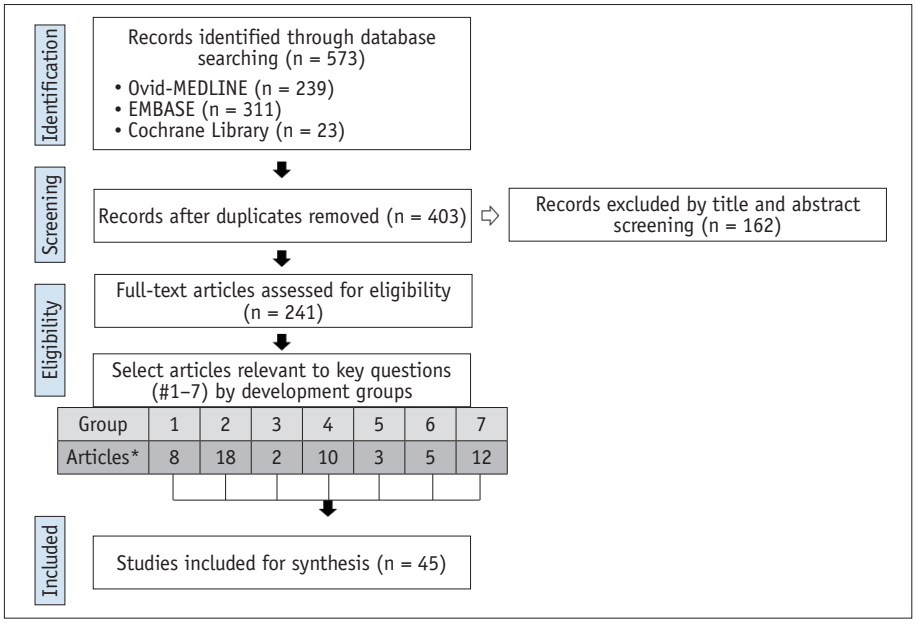

Fig. 1. Literature search was conducted using MEDLINE, Embase, and the Cochrane Library with keywords related to hepatocellular carcinoma, ultrasound, and Sonazoid. After applying exclusion criteria, 241 studies remained. Among the analyzed studies, 45 were selected by the development groups for their recommendations. *Numbers are total articles selected by each development group. They could be duplicated.

Table 1. Recommendation and Evidence Levels

Recommendation levels
Strongly recommended
Conditionally recommended
Evidence levels
I: Systematic review of cross-sectional studies with consistently applied reference standard and blinding
II: Individual cross sectional studies with consistently applied reference standard and blinding
III: Non-consecutive studies, or studies without consistently applied reference standards
IV: Case-control studies or poor or non-independent reference standard

Presentation and External Evaluation

The consensus recommendation statements were first presented at the KSAR annual conference on May 21, 2022. A draft of the guidelines was completed in January 2023 and subsequently reviewed for endorsement by both the KLCA and KSUM.

Sonazoid CEUS Examination Protocol

To propose a protocol for Sonazoid CEUS in HCC diagnosis, we reviewed the examination protocols of the studies in the evidence table (Supplementary Table 8). In most studies, the dose for a single injection of a suspension of Sonazoid powder and distilled water was 0.015 mL/kg (body weight), and a bolus injection was followed by a flush with normal saline. For scanning, a low mechanical index setting, from 0.2 to 0.3, was used.

The timing of vascular and Kupffer phase acquisition was examined: the vascular phase, which is the contrast enhancement of hepatic vessels that begins immediately after injection, was acquired up to 1 min or more after injection, and Kupffer phase, which is the delayed phase of microbubble ingestion by Kupffer cells, was acquired within 10 min after injection or later. In addition, reinjection in the Kupffer phase, which is the imaging used to confirm only the arterial enhancement of Kupffer-defective lesions, was performed. Among the 45 studies enrolled for guideline development, the vascular phase was obtained in 36 (80%), 21 of which analyzed the vascular phase for 1 min. The examination protocol for obtaining the vascular phase with Sonazoid is similar to that of other second-generation contrast agents [5]. The arterial phase begins when microbubbles appear in the hepatic artery (hepatic artery arrival, approximately 10 s after injection) and lasts for 20–35 s. The portal venous phase begins immediately at the end of the arterial phase (30–45 s after injection) and generally lasts for 2 min postinjection. Regarding the Kupffer phase, 32 studies (71%) obtained the Kupffer phase 10 min after injection, whereas 7 (16%) obtained it less

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than 10 min after injection or did not specify the timing. Six studies performed reinjections for the Kupffer phase washout. However, none of these studies have specifically defined the phase between the vascular and Kupffer phases. According to Shunichi et al. [6], the hepatic parenchyma is gradually enhanced from the start of vascular enhancement, and the enhancement curve of the parenchyma in the time-intensity curve intersects that of the portal vein at approximately 1 min. Therefore, they suggested that the portal venous phase ends after 60 s, called the succeeding period from 1 to 10 min the vasculo-Kupffer phase, and recognized the Kupffer phase at ≥ 10 min.

The presented guidelines suggest a Sonazoid CEUS examination protocol consisting of conventional vascular phases including the arterial phase (starts when microbubbles first arrive in the hepatic artery and ends when microbubbles fill the hepatic parenchyma), 1-min delay (early portal venous phase, ≤ 1 min from injection), and 2-min delay (late portal venous phase, ≤ 2 min from injection). The Kupffer phase, as the postvascular phase, was obtained with a delay of more than 10 min after injection. In addition, it is suggested that the time between the vascular and Kupffer phases be called the vasculo-Kupffer phase (transitional phase, from 2 to 10 min) (Fig. 2). Unlike the delayed phase of pure vascular agents, the vasculo-Kupffer phase reinforces the parenchymal enhancement. According to a study by Kang et al. [7], a 60-s cutoff for late washout and 6-min cutoff for the Kupffer phase showed the best diagnostic performance for HCC. Therefore, the clinical significance of the vasculo-Kupffer warrants further investigation.

Questions and Recommendations

A summary of the recommendations is presented in Table 2.

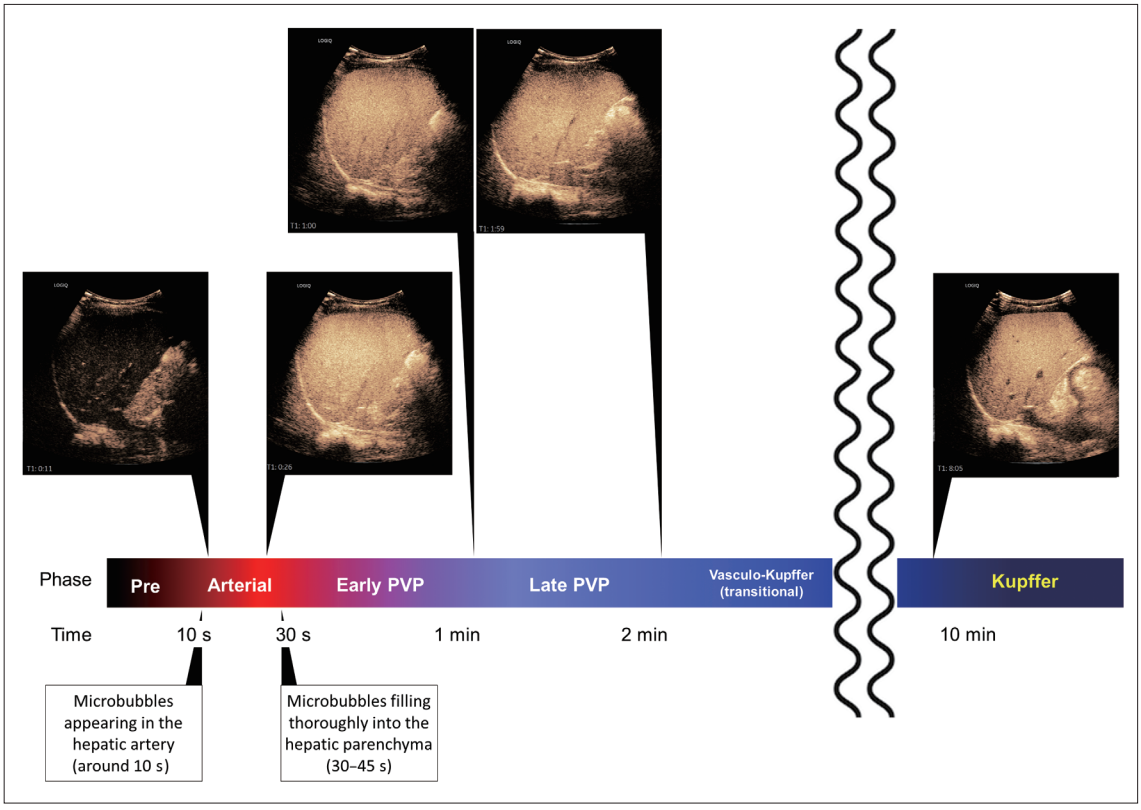

Fig. 2. Suggested imaging protocol for perfluorobutane contrast-enhanced ultrasound. Given the evidence, the following examination protocol is suggested: arterial phase (from the time when microbubbles appear in the hepatic artery to the start of the portal venous phase [PVP] [30–45 s]); PVP, divided into two sub-phases of early PVP (from the time when the liver parenchyma is completely filled with microbubbles [30–45 s] until 1 min later) and the late PVP (from 1 to 2 min after injection); the vasculo-Kupffer (transitional) phase (from 2 to 10 min after injection); and the Kupffer phase (≥ 10 min after injection). Analysis of video clips is important for evaluating the perfusion of hepatic lesions. These guidelines recommend recording a video clip (or frequent imaging capture) of the vascular phase from the start of the arterial phase to the end of the early PVP.

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Table 2. Summary of Recommendation Statements

Key Questions	Recommendations	Recommendation Level	Evidence Level	Agreement
1-1. Is it appropriate for nonrim APHE and late (≥ 60 s) and mild washout to be major imaging features of HCC in Sonazoid CEUS?	In Sonazoid CEUS, nonrim APHE and late (≥ 60 s) and mild washout are appropriate major imaging features for diagnosing HCC in at-risk patients.	Strongly	2	100% (18/18)
1-2. Can Kupffer phase washout be used as a major feature of HCC diagnosis in Sonazoid CEUS?	Kupffer phase washout can be considered another major imaging feature in Sonazoid CEUS for diagnosing HCC in at-risk patients if the lesions with nonrim APHE do not show either early or marked washout during the vascular phase.	Conditionally	2	100% (18/18)
2. What are the appropriate criteria for diagnosing HCC using Sonazoid CEUS in at-risk patients?	The diagnosis of HCC can be made in a nodule ≥ 1 cm detected in an at-risk patient when nonrim APHE with late and mild washout or washout in the Kupffer phase are present.	Conditionally	2	100% (18/18)
3. Can Sonazoid CEUS be used to characterize inconclusive nodules detected in CT or MRI in patients at high risk for HCC?	Sonazoid CEUS can be used to characterize nodules inconclusive in CT or MRI because it can detect the arterial hypervascularity of a nodule in real time and show Kupffer cell activity within the nodule.	Conditionally	3	88.9% (16/18)
4. Can Sonazoid CEUS differentiate HCC from non-HCC malignancies?	Sonazoid CEUS can differentiate HCC from non-HCC malignancies such as intrahepatic cholangiocarcinoma and metastasis.	Conditionally	3	88.9% (16/18)
5. Can Sonazoid CEUS be used as a surveillance tool for HCC in high-risk patients?	Sonazoid CEUS can be used as a surveillance tool in high-risk patients.	Conditionally	3	88.9% (16/18)
6. Is Sonazoid CEUS helpful for guiding local ablation therapy for HCC?	Sonazoid CEUS is helpful for guiding local ablation therapy because it increases the detectability and conspicuity of small HCCs that are inconspicuous on B-mode ultrasound.	Strongly	2	100% (18/18)
7. Is it appropriate to use Sonazoid CEUS to assess the treatment response of HCC in patients who underwent TACE or RFA?	When less than three index tumors can be observed with CEUS, Sonazoid CEUS may be useful for evaluating treatment response after TACE or RFA.	Conditionally	3	94.5% (17/18)

APHE = arterial phase hyperenhancement, CEUS = contrast-enhanced ultrasonography, HCC = hepatocellular carcinoma, CT = computed tomography, MRI = magnetic resonance imaging, TACE = transarterial chemoembolization, RFA = radiofrequency ablation

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Question 1. What are the Major Features of HCC in Sonazoid CEUS?

Question 1-1. Is it Appropriate for Nonrim APHE and Late (≥ 60 s) and Mild Washout to be Major Imaging Features of HCC in Sonazoid CEUS?

[Recommendation]

In Sonazoid CEUS, nonrim APHE and late (≥ 60 s) and mild washout are appropriate major imaging features for diagnosing HCC in at-risk patients (**Recommendation level: strongly recommended, Evidence level II**).

During hepatocarcinogenesis, the density of the unpaired arteries progressively increases [8]. Therefore, more enhancement in the arterial phase than in the liver without peripheral rim enhancement on computed tomography (CT)/magnetic resonance imaging (MRI) (nonrim APHE) is a major imaging feature for diagnosing HCC in at-risk patients [9-12]. Similarly, APHE on Sonazoid CEUS is an important imaging feature for diagnosing HCC in at-risk patients. As a prerequisite, rim APHE, spokewheel, centrifugal APHE, and peripheral discontinuous nodular APHE should not be considered as major findings of HCC on Sonazoid CEUS. This is because rim APHE can be observed in other malignancies, including intrahepatic cholangiocarcinoma (ICC); spokewheel and centrifugal APHE are suggestive of focal nodular hyperplasia; and peripheral discontinuous nodular APHE is indicative of hepatic hemangioma. Diffuse APHE, also known as nonrim APHE, is a key finding in HCC. According to a previous study comparing CT/MRI LI-RADS with Sonazoid CEUS in patients with suspected HCC [13], 89.6% of CT/MRI LR-5 (definitely HCC) and 85.9% of LR-4 (probably HCC) patients showed nonrim APHE on Sonazoid CEUS, compared with 57.6% of LR-3 (intermediate probability of malignancy) patients. In a Sonazoid CEUS study of 113 at-risk nodules [7], the presence of nonrim APHE was more frequent in HCC (86.8% [72/82]) than in non-HCC malignancies (56.2% [9/16]) and benign lesions (20% [3/15]). Consistent results were also found in a study of 59 at-risk nodules [14]: nonrim APHE was observed in 95% (41/43) of HCCs, 30% (3/10) of non-HCC malignancies, and 16.7% (1/6) of benign lesions. In addition, in an intraindividual comparison of pure blood-pool CEUS and Sonazoid CEUS [14], the enhancement patterns of HCC in the arterial phase were the same, perhaps because the arterial phase of Sonazoid CEUS is a nearly pure vascular image with negligible effects on Kupffer cell uptake.

Another key change during hepatocarcinogenesis is the diminishing portal triad [8]. This results in a washout appearance, defined as a temporal reduction in enhancement relative to the composite liver tissue from earlier to later phases [10]. Thus, washout on CT/MRI is a major imaging feature for the diagnosis of HCC in at-risk patients [9-12]. Unlike CT/MRI, early CEUS studies reported that washout appearance was insufficient to differentiate HCC from ICC [15]. However, succeeding studies have revealed that the late (≥ 60 s) timing and mild (when the nodule enhances less than the liver but not devoid of enhancement) degree of washout are important factors in differentiating HCC from other malignancies in CEUS examination using a blood-pool agent [16-20]. However, the unique characteristic of Sonazoid (taken up by Kupffer cells) requires a delicate approach to assess washout because it may cause a pseudo-washout effect. In a study that observed washout timing from 50 s after Sonazoid injection, 21% of HCC (15/73) showed washout before 60 s, although the most frequent timing for the washout to start was 72–120 s. Nevertheless, the specificity and positive predictive value (PPV) were 100% when late washout was defined as 60 s on Sonazoid CEUS [7].

Therefore, in Sonazoid CEUS, nonrim APHE and late and mild washout are appropriate major imaging features for diagnosing HCC in at-risk patients; this recommendation is supported by four studies [7,13,14,21].

Question 1-2. Can Kupffer Phase Washout be Used as a Major Feature of HCC Diagnosis in Sonazoid CEUS?

[Recommendation]

Kupffer phase washout can be considered another major imaging feature in Sonazoid CEUS for diagnosing HCC in at-risk patients if lesions with nonrim APHE do not show either early or marked washout during the vascular phase (**Recommendation level: conditionally recommended, Evidence level II**).

Kupffer phase washout is defined as a hypo-enhancing area compared with the liver in the Kupffer phase and reflects a decreased number of Kupffer cells. The advantage of considering the Kupffer phase washout as a major imaging feature in Sonazoid CEUS is the improved sensitivity of HCC diagnosis in at-risk patients. According to previous

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Sonazoid CEUS studies, 7.8%–13.0% of histopathologically proven HCCs with nonrim APHE showed washout only in the Kupffer phase and not in the late vascular phase [22-26]. Therefore, Kupffer phase washout has the potential to diagnose additional cases of HCC compared with late and mild washout. The nonrim APHE with washout only in the Kupffer phase were more common in early HCCs (4/16 [25%]), nodule-in-nodule type HCCs (5/5 [100%]), and well-differentiated HCCs (3/16 [18.8%]) than in overt HCCs (0/48) or moderately and poorly differentiated HCCs (3/42 [7.1%] and 0/6, respectively) [22,23]. In another prospective study of at-risk patients, Kupffer phase washout (93%) was a more frequent Sonazoid CEUS finding of HCC than were late (≥ 60 s) and mild washout (77%–79%) [14]. When the modified LR-5 criteria, defined as nonrim APHE without early washout followed by Kupffer phase washout, were applied to at-risk patients, a high PPV of 93.8% for HCC diagnosis was reported [27].

However, because Kupffer phase washout is not specific for HCC, there is a risk of reduced specificity. Therefore, exceptions, such as the exclusion of early or marked washout during the vascular phase (similar to the CEUS LI-RADS), are required when assessing Kupffer phase washout [28]. The results of previous Sonazoid CEUS studies using histopathologically proven malignant lesions revealed that all metastases (9/9), 91.3% of ICC (21/23), and 80% of combined hepatocellular cholangiocarcinoma (4/5) showed rim APHE, early (< 60 s) washout, and/or marked (when the nodule lacks of any contrast enhancement within 2 min after contrast injection; in other words "punched-out") washout [14,24,27]. In a study of the diagnostic performance of Sonazoid CEUS in at-risk patients comparing the LI-RADS criteria (defined as nonrim APHE and late [≥ 60 s]/mild washout) with the nonrim APHE and Kupffer criteria (defined as nonrim APHE and Kupffer phase washout), the nonrim APHE and Kupffer criteria provided higher sensitivity and accuracy for HCC diagnosis without loss of specificity [24]. Therefore, in at-risk patients, Kupffer phase washout can be considered a major imaging feature for diagnosing HCC on Sonazoid CEUS if the observations show no rim APHE or early or marked washout during the vascular phase.

Question 2. What are the Appropriate Criteria for Diagnosing HCC Using Sonazoid CEUS in At-Risk Patients?

[Recommendation]

The diagnosis of HCC can be made in a nodule ≥ 1 cm detected in an at-risk patient when nonrim APHE with late and mild washout or washout in the Kupffer phase are present (**Recommendation level: conditionally recommended, Evidence level II**).

Nineteen studies reported the performance of Sonazoid CEUS in the diagnosis of HCC [13,14,27,29-44]. These studies have the following limitations: 1) 68% (13/19) were retrospective [13,27,29-32,34-37,39,40,42], and 2) detailed diagnostic criteria for HCC were not clearly described. Most studies claimed that they used nonrim APHE and Kupffer phase washout as HCC diagnostic criteria in Sonazoid CEUS, with or without the combined use of late and mild washout during the vascular phase. However, the definition of the degree or timing of washout was not explicitly presented, and some studies were ambiguous regarding how they combined late and mild washout and Kupffer phase washout. Therefore, precise and predefined criteria and terminology, such as the CEUS LI-RADS, should be applied in future studies.

A prospective study comparing Sonazoid and pure blood-pool CEUS intraindividually adopted the CEUS LI-RADS diagnostic criteria for HCC, that is, nonrim APHE with late and mild washout in a nodule ≥ 1 cm. Sonazoid CEUS showed a significantly higher sensitivity (79%) than pure blood-pool CEUS (54%) and the same specificity (100%) [14]. Therefore, we conclude that the CEUS LI-RADS diagnostic criteria for HCC could be applied to Sonazoid CEUS.

Given that the CEUS LI-RADS diagnostic criteria for HCC were originally developed for blood-pool agents, modifications may be necessary when applying these criteria to Sonazoid CEUS. In a recent retrospective study, the modified CEUS LI-RADS HCC diagnostic criteria using Kupffer phase washout outperformed conventional mild and late washout in terms of sensitivity (modified criteria vs. conventional criteria, 83% vs. 74%) without a significant loss of specificity (64% vs. 70%) [13]. Another retrospective study reported a sensitivity and specificity of 70% and 93%, respectively, using the same modification as the CEUS LI-

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RADS HCC diagnostic criteria [27]. These studies suggest that the modified CEUS LI-RADS HCC criteria embracing the Kupffer phase washout in Sonazoid CEUS may be useful.

Accordingly, in these guidelines, we propose that the diagnosis of HCC can be made in a nodule ≥ 1 cm in at-risk patients when nonrim APHE is present with late and mild washout or Kupffer phase washout. To maintain specificity, Kupffer phase washout in HCC can be applied only when lesions do not show either early washout (< 60 s) or marked washout during the vascular phase because non-HCC malignancies can show Kupffer phase washout [45]. Similar to the CEUS LI-RADS criteria, APHE should not be applied to lesions with a rim or peripheral discontinuous nodular enhancement during the arterial phase, which are typical imaging features of ICC and hemangioma, respectively [45]. Given that several cross-sectional studies [13,27] have reported the performance of the modified CEUS LI-RADS HCC diagnostic criteria with a proper study design, these diagnostic criteria can be conditionally recommended (level 2).

Question 3. Can Sonazoid CEUS be Used to Characterize Inconclusive Nodules Detected in CT or MRI in Patients at High Risk of HCC?

[Recommendation]

Sonazoid CEUS can be used to characterize inconclusive nodules on CT or MRI because it can detect the arterial hypervascularity of a nodule in real time and show Kupffer cell activity within the nodule (**Recommendation level: conditionally recommended, Evidence level III**).

Unlike other cross-sectional imaging modalities, such as contrast-enhanced CT or MRI, which can evaluate the whole liver, Sonazoid CEUS is generally performed to examine a small portion of the liver. Therefore, the Asian Pacific Association of the Study of the Liver and the KLCA-National Cancer Center Korea practice guidelines recommend Sonazoid CEUS as a second-line imaging modality for nodules inconclusive on CT and MRI [46,47]. The advantages of Sonazoid CEUS over contrast-enhanced CT or MRI include its excellent temporal resolution. Contrast-enhanced CT or MRI can acquire arterial-phase images at a single time or a few time points, whereas Sonazoid CEUS enables real-time monitoring of the liver during the early vascular phase. Therefore, Sonazoid CEUS has the potential to detect arterial hypervascularity that is missed on contrast-enhanced CT or MRI because of a limited time window or inappropriate timing. According to previous studies, Sonazoid CEUS detected arterial hypervascularity in 29.4% and 43.2% of lesions that did not demonstrate hypervascularity on contrast-enhanced CT and MRI, respectively [48,49].

Another advantage of Sonazoid CEUS over contrast-enhanced CT or MRI is the ability to acquire Kupffer phase images. From a safety viewpoint, Sonazoid CEUS is free from radiation hazard and considered to be safe [50]. Therefore, Sonazoid CEUS can be used to characterize inconclusive nodules on contrast-enhanced CT or MRI owing to its excellent temporal resolution and Kupffer phase imaging.

Question 4. Can Sonazoid CEUS Differentiate HCC from Non-HCC Malignancies?

[Recommendation]

Sonazoid CEUS can differentiate HCC from non-HCC malignancies such as intrahepatic cholangiocarcinoma and metastasis (**Recommendation level: conditionally recommended, Evidence level III**)

ICC is the second most common primary hepatic malignancy after HCC and is occasionally detected during the surveillance of patients at risk of HCC [51]. Therefore, accurate differentiation between ICC and HCC is of utmost importance, especially in the presence of cirrhosis. On B-mode ultrasound, ICC manifests as a mass of varying echogenicity, depending on its size, and is usually accompanied by irregular margins, a peripheral hypoechoic rim, and dilation of the peripheral bile ducts [52].

The typical (common) features of ICC on pure blood-pool CEUS are rim APHE, early washout, and marked washout [17,32,53-55], also referred to as LR-M on CEUS LI-RADS [28]. The feature exclusively available on Sonazoid CEUS, marked by Kupffer phase washout 10 min after contrast injection, showed a high PPV for diagnosing non-HCC malignancies [27]. Regarding rim APHE on Sonazoid CEUS in patients at risk of HCC, a recent study reported that the sensitivity in differentiating ICC from HCC was suboptimal (16.7%, 1/6), although the specificity was 100% (56/56) [27]. Similarly, Kang et al. [14] found that the sensitivity of rim APHE for diagnosing non-HCC malignancies ranged from 40% to 50%; however, its specificity was 100% (43/43). Early or marked washout appears to be more useful than rim APHE for

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differentiating ICC from HCC on Sonazoid CEUS, with high sensitivity (100%, 6/6) and specificity (90.6%, 58/64) in at-risk patients [27]. This washout pattern reflects the low blood volume and large extracellular interstitial space of the ICC [15,56,57], and especially, the degree of washout is more prominent in Sonazoid CEUS due to Kupffer cell uptake in surrounding normal tissues. This pattern differs from the overall unwashed gradual enhancement commonly observed on CT and MRI. Unlike the microbubbles used for CEUS, the contrast agents used in CT/MRI can leak through the vascular endothelium and accumulate in the tissue interstitium, which causes the absence of the washout observed with ICCs on CT/MRI [56,58-60]. In addition, some subsets of HCC, such as poorly differentiated HCC and HCC with microvascular invasion, may show early washout on CEUS [61,62]. Conflicting results have been reported regarding the diagnostic value of marked Kupffer phase washout. One study reported that all ICCs had marked Kupffer phase washout (6/6); however, only 25.0% (16/64) of HCCs showed marked Kupffer phase washout [27]. In contrast, another study reported that the degree and prevalence of Kupffer phase washout did not differ between the two diseases [14]. Therefore, further studies are required to reveal the role of marked hypoenhancement in the Kupffer phase in differentiating ICC from HCC.

The differential diagnosis of metastasis from HCC is less important than that from ICC, even though HCC is the most common hepatic malignancy in non-cirrhotic livers. Nevertheless, the imaging characteristics of metastasis on Sonazoid CEUS are well known: rim APHE, hypo-enhancement during the portal venous and vasculo-Kupffer phases, and a clearly demarcated hypoechoic defect in the Kupffer phase because such lesions do not contain Kupffer cells [5,31,32,63-65].

Overall, Sonazoid CEUS shows sufficient diagnostic performance with high sensitivity and specificity for the differential diagnosis of focal liver lesions [32,36,37,44,65]. However, given the overlapping imaging features, including rim APHE and early washout among the subsets of HCC, ICC, and metastasis, a clear CEUS-based diagnosis may be limited.

Question 5. Can Sonazoid CEUS be Used as a Surveillance Tool for HCC in High-Risk Patients?

[Recommendation]

Sonazoid CEUS can be used as a surveillance tool in high-risk patients (**Recommendation level: conditionally recommended, Evidence level III**).

B-mode ultrasonography is widely used as the primary imaging modality for HCC surveillance in high-risk patients. However, it is substantially less effective in detecting small HCCs in patients with cirrhosis who exhibit severe architectural distortion or an extremely poor sonic window [66]. Sonazoid CEUS has been investigated for its potential to overcome the limitations of B-mode ultrasound by providing a long and stable Kupffer phase, which makes it possible to detect HCCs that present as Kupffer phase washout throughout the liver. Therefore, Sonazoid CEUS is expected to be particularly sensitive in detecting small HCCs in at-risk patients, especially in those with coarsened hepatic echotexture.

In an early study by Kudo et al. [67], Sonazoid CEUS identified 27 Kupffer phase washout lesions that were not detected on B-mode ultrasonography in 292 patients with cirrhosis. In 16 of these 27 lesions, tumor hypervascularity was confirmed using the defect-reperfusion imaging technique, and all were histologically proven to be HCC (size range, 6–13 mm). In another study, nine additional nodules, including seven HCCs, were detected on Kupffer phase imaging in 262 patients with cirrhosis [68]. A multicenter randomized controlled trial compared Sonazoid CEUS Kupffer phase surveillance with B-mode ultrasound surveillance in very high-risk Japanese patients and found that the mean size of HCCs at first detection was significantly smaller in the Kupffer phase group than in the B-mode ultrasound group (13.0 vs. 16.7 mm, $P = 0.011$) [69]. Interestingly, a subgroup analysis found this difference in patients with hepatitis C but not in patients with hepatitis B. Another recent prospective multicenter intraindividual comparison study in Korean patients with a predominance of hepatitis B (SCAN trial) demonstrated that adding Sonazoid CEUS to B-mode ultrasound during HCC surveillance slightly increased the detection rate for early-stage HCC (0.8% vs. 1.1%, $P = 0.160$) and significantly reduced the false referral rate (4.4% vs. 1.1%, $P < 0.001$) [70]. Although no protocol has yet been established, when Sonazoid CEUS is used for

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surveillance, it may be more appropriate to use only Kupffer phase imaging without vascular phase imaging to minimize examination time, especially in high-volume centers. In this setting, if a lesion with Kupffer phase washout is detected, a defect reperfusion study can be performed to assess hypervascularity with the additional administration of Sonazoid [2]. In contrast, a full Sonazoid CEUS examination can be considered if CT or MRI access is limited or if the patient has renal dysfunction or hypersensitivity to CT or MRI contrast agents.

Cost-effectiveness is an important factor in determining whether a modality can be used as a surveillance tool. Only one Japanese study has considered this aspect of Sonazoid CEUS [71]. Compared with the no surveillance group, the B-mode ultrasound and Sonazoid CEUS surveillance groups showed an incremental cost-effectiveness ratio (ICER) of $17296 US dollars (USD)/quality-adjusted life-year (QALY) and $18384 USD/QALY, respectively, which were below the commonly accepted threshold of $50000 USD/QALY. Furthermore, when the Sonazoid CEUS group was compared with the B-mode ultrasound group, the ICER was $24250 USD; therefore, Sonazoid CEUS was cost-effective. However, that study had insufficient generalizability because it used a decision-making model based on the natural history of chronic hepatitis C and cost data based only on the literature published in Japan.

Question 6. Is Sonazoid CEUS Helpful for Guiding Local Ablation Therapy for HCC?

[Recommendation]

Sonazoid CEUS is helpful for guiding local ablation therapy because it increases the detectability and conspicuity of small HCCs that are inconspicuous on B-mode ultrasound (**Recommendation level: strongly recommended, Evidence level II**).

Generally, local ablation therapy for HCC is performed under ultrasound guidance owing to its convenience and real-time capability. Recent advanced MRI techniques, such as hepatocyte-specific contrast agents and diffusion-weighted imaging, can diagnose very small HCCs [72,73]. However, locating and treating such small HCCs can be challenging because they tend to have poor conspicuity on B-mode ultrasound [74]. The unique characteristic of Sonazoid CEUS, Kupffer phase imaging, increases the sensitivity of HCCs by means of the echogenicity difference between the target lesion and normal liver parenchyma [2]. Furthermore, the Kupffer phase lasts for 60–120 min after intravenous injection of Sonazoid [24], and a long time window enables local ablation therapy for small HCCs.

Many studies have demonstrated the strength of Sonazoid CEUS-guided radiofrequency ablation (RFA) over B-mode ultrasound-guided procedures based on improving lesion detectability and decreasing the number of sessions of local ablation therapy required to achieve efficacy [39,75-77]. According to a previous study, Sonazoid CEUS found 69 more nodules in 52 patients than B-mode ultrasound alone. The detection rates of HCC using Sonazoid CEUS and B-mode ultrasound were 93.2% and 83.5%, respectively (P = 0.04) [39]. A prospective study by Lee et al. [76] reported that Kupffer phase imaging increased lesion conspicuity and the operator's diagnostic confidence in 29 patients with 31 HCCs (31/43, 72%) compared with B-mode ultrasound, in which the HCCs were poorly identifiable from the surrounding cirrhosis-related nontarget lesions.

Sonazoid CEUS guidance can reduce the number of treatment sessions needed to achieve efficacy compared with the number required under B-mode ultrasound guidance alone [39,77]. Sonazoid CEUS guidance can also lead to better therapeutic outcomes than B-mode ultrasound guidance through sustained local tumor control [78]. According to a study by Minami et al. [77], the sustained local control rate was higher in the CEUS-guided RFA group than in the B-mode ultrasound-guided group (92.1% vs. 76.3% and 85.3% vs. 66.4% at 1 and 2 years, respectively).

Ultrasound fusion imaging with CT or MRI is commonly used to locate index tumors during local ablation therapy for HCC. Fusion imaging with Sonazoid CEUS can be a powerful tool to improve lesion conspicuity and technical feasibility of local ablation therapy for HCC [79].

Question 7. Is it Appropriate to Use Sonazoid CEUS to Assess the Treatment Response of HCC in Patients who Underwent Transarterial Chemoembolization (TACE) or RFA?

[Recommendation]

When fewer than three index tumors can be observed with CEUS, Sonazoid CEUS may be useful for evaluating treatment response after TACE or RFA (**Recommendation level: conditionally recommended, Evidence level III**).

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Generally, contrast-enhanced CT or MRI is used to evaluate the treatment response of HCC, and its effectiveness has been verified. Although its application is relatively limited, several studies have tested the use of Sonazoid CEUS to evaluate HCC response after locoregional treatment [80-82]. In patients who underwent ultrasound-guided RFA, Sonazoid CEUS was used to assess the viability of HCC 3 h after the procedure if additional ablation was needed [81]. In another study, CEUS using Sonazoid was more sensitive than contrast-enhanced CT for evaluating a margin of 5 mm after RFA for HCC [80]. Furthermore, after RFA and TACE for HCC, serial CEUS follow-up using Sonazoid was more accurate in diagnosing local recurrence and less affected by observer experience than dynamic CT [83]. Moreover, for a few index tumors observed using CEUS, Sonazoid CEUS was helpful in evaluating HCC viability or response after TACE or transarterial radioembolization [84-86]. However, because the number of patients was not large enough in most studies and there have been no randomized controlled trials, clinical evidence is insufficient to make a strong recommendation.

Several studies have shown that Sonazoid CEUS and perfusion parameters are useful in predicting treatment responses after radiotherapy and systemic therapy. Funaoka et al. [87] reported that Sonazoid CEUS was helpful for evaluating HCC after radiation treatment. After treating HCC with systemic agents such as sorafenib, Sonazoid CEUS was helpful in predicting treatment response by examining perfusion parameters or evaluating enhancement architecture [88-91]. However, most studies considered fewer than 50 patients and had preliminary study designs in which evaluation parameters were not established. Furthermore, it is difficult to recommend Sonazoid CEUS as a modality for evaluating HCC after radiotherapy or systemic treatment because a perfusion parameter or enhancement pattern evaluation using CEUS is complicated to apply in clinical practice. Sonazoid CEUS could be a supplementary option for evaluating the response of HCC when a few index tumors are observed simultaneously.

CONCLUSION

With an increasing number of countries approving the use of Sonazoid CEUS for liver lesions, the number of published studies is increasing, creating a need for guidelines that include the diagnostic criteria for HCC using Sonazoid CEUS. This guideline was developed by collecting as much literature as possible on Sonazoid CEUS, followed by an in-depth review by experts in the field and a fair consensus process to help healthcare providers make clinical decisions about performing Sonazoid CEUS on patients at risk of HCC and utilize the results to guide the treatment of HCC.

Supplement

The Supplement is available with this article at https://doi.org/10.3348/kjr.2023.0324.

Conflicts of Interest

Min Woo Lee, Jung Hoon Kim, Ijin Joo, So Yeon Kim, Yong Eun Chung, Jeong Min Lee, contributing editors of the *Korean Journal of Radiology*, were not involved in the editorial evaluation or decision to publish this article. Woo Kyoung Jeong received honorarium from GE Healthcare and Bayer; Hyo-Jin Kang received honorarium from GE Healthcare; Mi-Suk Park received grant from GE Healthcare and Guerbet; Bohyun Kim was a consultant for Samsung Medison; Min Woo Lee received honorarium from Bracco, Starmed, Bonston Scientific, and Medtronic, and was a consultant for Starmed and Medtronic; Jeong Ah Hwang received honorarium from GE Healthcare; Jae Young Lee received grant from Canon Healthcare, Siemens Healthcare, Alpinion Medical System, and GE Healthcare and received honorarium from Philips, Canon Healthcare, GE Healthcare, Alpinion Medical System, and Siemens Healthcare; Jung Hoon Kim received honorarium from GE Healthcare; Ijin Joo received honorarium from Samsung Medison; So Yeon Kim received grant from Bayer Healthcare and Canon Healthcare; Jeong Min Lee received grant from Bayer Healthcare, Canon Healthcare, Siemens Healthcare, GE Healthcare, CMS, Guerbet, Samsung Medison, Starmed, RF medical, Clarify, and Dongkuk Pharma and received honorarium from Samsung Medison, Philips, GE Healthcare, Bayer, Guerbet, and Clarify. The funders had no role in the data analysis, or the decision to publish. All remaining authors have declared no conflicts of interest.

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