Cancer Imaging (2005) **5,** 39–45
DOI: 10.1102/1470-7330.2005.0011

CI

e-med
www.e-med.org.uk

ARTICLE

Role of contrast-enhanced color Doppler ultrasonography and dynamic flow in the evaluation of hepatic tumors treated with radiofrequency

N Lassau*, M Lamuraglia*†‡, I Chawi*, T Smayra*, C Dromain*, S Koscielny†, T de Baere*, J Leclère* and A Roche*

**Department of Medical Imaging, Institut Gustave Roussy, 39 rue Camille Demoulins, 94805 Villejuif, France; †Department of Biostatistics, Institut Gustave Roussy, 94805 Villejuif, France; ‡Section of Internal Medicine, Department of Internal Medicine and Public Medicine, University of Bari, Piazza Giulio Cesare 11, 70124 Bari, Italy*

Corresponding address: Nathalie Lassau, 39 rue Camille Demoulins, 94805 Villejuif Cedex, France
E-mail: lassau@igr.fr

Date accepted for publication 13 April 2005

Abstract

The contribution of contrast-enhanced color Doppler ultrasonography (CDUS) and dynamic flow (DF) (Toshiba) in the evaluation following treatment of hepatic tumors with radiofrequency (RF) is discussed. Twenty-seven patients with 34 hepatic tumors were included in this prospective study. The treated tumors measured 10–58 mm in diameter (mean diameter 29 mm). Two tumors were treated twice and one three times, comprising a total of 38 target lesions treated with RF and evaluated by 127 contrast-enhanced CDUS. The results of CDUS follow-up were compared to those of the dynamic MRI at 2 months, 4 months, 6 months and 1 year. Before RF, the injection of Levovist raised the number of vascularized lesions seen with unenhanced Doppler from 44% to 79%. All the non-vascularized lesions were metastases. Twenty-four hours after RF, four tumors presented an enhancement with Levovist, in which two were insufficiently treated lesions. Twenty-one treated tumors have been followed-up jointly by CDUS and MRI at the same time at 2 months, 20 at 4 months, 12 at 6 months and nine at 1 year. Compared to the MRI and the evolution, the CDUS presented a sensitivity of 100% and a specificity of 90% for the detection of progressive recurrence. The preliminary results show that the CDUS is useful to confirm the absence of detectable vascularity after treatment with RF ablation, whereas the presence of enhancement must be confirmed by MRI.

Keywords: *Hepatic tumors; radiofrequency ablation; ultrasound; contrast media; Doppler studies; magnetic resonance.*

Introduction

Thermal ablation by radiofrequency (RF) has become a widely used technique for treatment of hepatic metastases and hepatocellular carcinoma (HCC)[1–3]. With this technique the induced lesion constitutes a generally spherical area, which presents a coagulation necrosis replacing the treated tumor where histopathological findings lack any viable tissue or any vascular structure. The published series show that it is a very effective technique on a short-term basis but its long-term success relies on detection and early treatment of local recurrence[4], which needs a reliable, available and easy to perform follow-up technique. The actual methods of follow-up are based on computed tomography (CT), and more recently on contrast-enhanced magnetic resonance imaging (MRI) with dynamic acquisitions[5,6]. Several groups have also become interested in the potential role of contrast-enhanced color Doppler ultrasound (CDUS)[7,8] in the assessment of the immediate efficiency of RF, and the detection of early signs of recurrence. Our aim was to

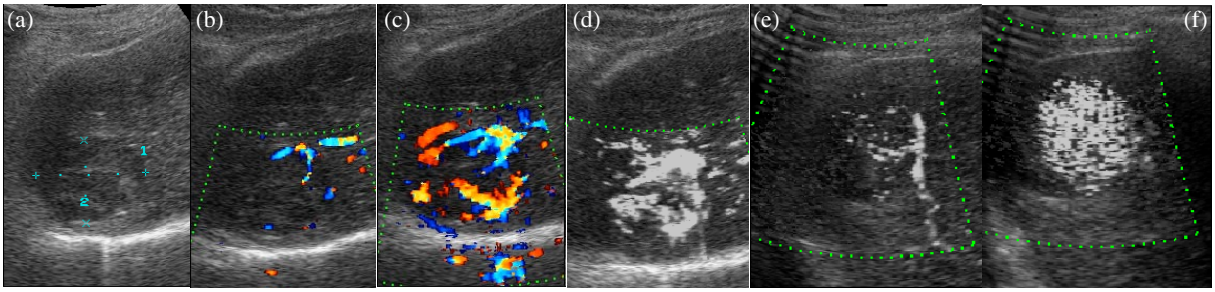

Figure 1 **A 71-year-old patient with HCC lesion. Assessment before treatment: (a) Hypoechoic 38 mm liver tumor on mode B. (b) Color Doppler study without Levovist shows a moderate peripheral vascularization. (c) Contrast-enhanced color Doppler study shows a clear intratumoral enhancement. However, a blooming effect persists. (d) Contrast-enhanced dynamic flow Doppler demonstrates vascular and parenchymal enhancement of the tumor. (e) Precocious phase of injection of Levovist. (f) Late (parenchymal) phase with Levovist.**

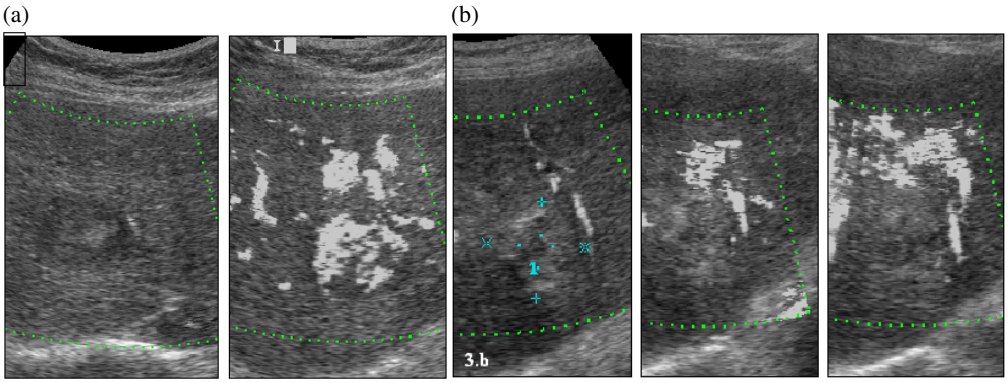

Figure 2 **CDUS examination 24 h after RF ablation in a 70-year-old patient with HCC: (a) Before RF: heterogeneous hypoechoic 34 mm lesion. CDUS shows important intratumoral enhancement. (b) After RF: 40 mm hyperechoic heterogeneous treated lesion with enhancement only in the adjacent normal tissue; no enhancement in the treated area.**

describe a method of follow-up by CDUS and to present our preliminary results.

Materials and methods

In this prospective study, we intended to include 40 patients with hepatic tumors (HCC or metastases) treated with percutaneous radiofrequency ablation (RF). This protocol has obtained the agreement of the ethical committee of our institute. All patients provided informed consent.

Twenty-seven patients with 34 lesions have been included in the study (15 women and 12 men; 40–79 years old, mean age 60 years). Eight patients had HCC and 19 had metastases. Two tumors underwent RF ablation twice and one three times. In total, 38 sessions of RF were carried out for 38 lesions (10 HCC, 28 metastases) of which four were recurrences.

To these, 33 tumor targets were performed before treatment with RF, 22 controls in the 24 h after treatment, 23 CDUS evaluation at 2 months, 20 at 4 months, 14 at 6 months and 10 at 1 year, with a total of 127 contrast-enhanced CDUS. The results of 67 follow-up CDUS were compared to those of the 62 corresponding MRI studies at 2 months, 4 months, 6 months and 1 year, and to the clinical evolution (Table 1).

Table 1 ***Evaluation time of lesions with CDUS and MRI***

	Evaluation time					
	D − 1	D + 1	2 months	4 months	6 months	1 year
CDUS (no. of lesions)	33	22	23	20	14	10
MRI (no. of lesions)			21	20	12	9

Two sonographs were used. At first we used an AU4 (Escoate Biomedica, Genoa, Italy) with 3.5 MHz transducer. Next we used a Powervision 8000 (Toshiba, Japan) with a C37 convex-array transducer maintained at 4.4 MHz with dynamic flow perfusion software (DF) which allowed, thanks to broadband Doppler technology, a flow imaging of excellent spatial resolution, with elevated image rate and suppression of blooming effect.

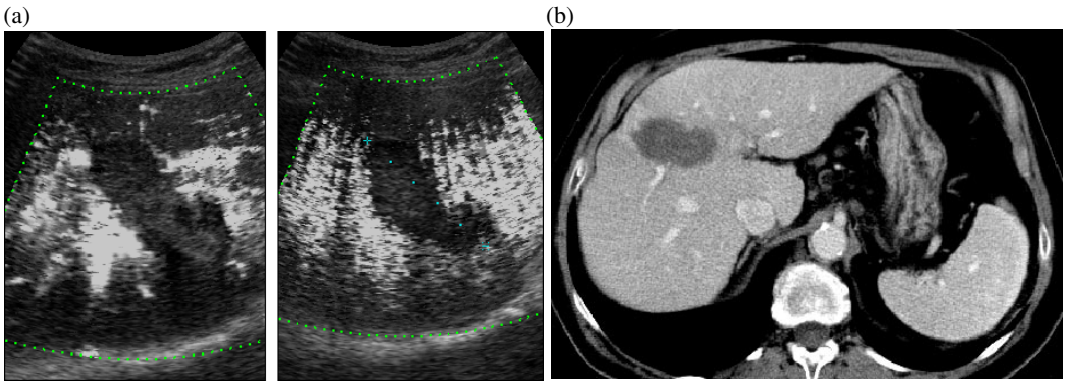

Figure 3 **A 69-year-old patient with HCC. Correlation between CDUS and CT at 4 months after RF ablation: (a) CDUS shows parenchymal enhancement in the normal hepatic tissue with no enhancement in the treated area. (b) No enhancement in the treated area.**

Patients were placed in supine or left lateral decubitus position to optimize the sonographic evaluation of the target lesion. Each CDUS was performed in four steps:

1. *Morphological study* was performed in B mode, allowing the identification of the target lesion, the best acoustic window to assess it and assessment of respiratory movements of the patient. This window has been maintained as far as possible for all the examinations of the same patient. For each lesion, the largest diameter was measured with callipers; the echogenicity and the heterogeneous or homogenous aspects were noted.

2. *Doppler study before IV injection*: for this second step, CDUS was used for the entire examination with sensitivity adjustment adapted to slow flow (pulse repetition frequency 0.5–4.5 kHz) and for the most recent examinations using dynamic flow (DF). The number of visible vessels inside the tumor was counted and classified into three groups: no vessels, 1–4 vessels, and more than 4 vessels.

3. *Injection of sonographic contrast agent*: Levovist (SH U 508 A, Shering, Berlin, Germany) is a suspension of micrometer-sized microparticles of galactose and microscopic gaseous bubbles in sterilized water prepared by agitation of 4 g of microparticles in sterilized water for 10 s and left to settle for 2 min. This gives a suspension of 10 ml at a concentration of 400 mg/ml; it must be injected intravenously at 10 ml/s.

4. *Dynamic study after injection of Levovist* (contrast-enhanced CDUS): the intravascular signal enhancement of the intratumoral neovessels after injection was evaluated visually in real time and the dynamic sequence was recorded on videotape (U-matic) for the earlier examinations and subsequently on a digital tape. We noted the time of injection, the time of the start of enhancement and the maximum enhancement. Every subjective or objective reaction was also noted.

With DF, the emission power was chosen to obtain an elevated mechanic index (MI), necessary to explode the Levovist microbubbles and to enhance the signal of all vascularized tissue. The sweeping was stopped for 3–5 s almost every 10 s to allow the patient to breathe and the microbubbles to further accumulate in tiny residual tumoral tissues that are then revealed at the resumption of sweeping by signal enhancement due to the explosion of accumulated microbubbles.

Two radiologists reviewed the color Doppler (CD) examinations for the number of vessels, seen as intratumoral colored dots or areas; the DF were examined for the number of vessels and the presence or absence of intratumoral ‘parenchymography’ supposed to represent the remaining viable tumoral tissue.

MR imaging was performed with a 1.5-T whole-body MR imager (Signa LX; GE Medical Systems). All MR images were obtained in the transverse plane with a phased-array multicoil for the body. Section thickness was 7 mm, with a 2 mm intersection gap for all pulse sequences. The imaging protocol comprised fat suppressed T2-weighted respiratory-triggered fast spin-echo sequences. Dynamic contrast material-enhanced MR imaging was performed at four consecutive 30 s intervals and 5 min after the start of a bolus injection of 0.1 mmol/kg of body weight of gadoterate meglumine (Dotarem; Guerbet) into the antecubital vein using a power injector (Spectris; Medrad). T1-weighted fast multiplanar spoiled gradient-recalled echo (GRE) sequences were performed.

Results

Treated tumors measured 10–58 mm (mean 29 mm). Tumoral echostructure and appearance were not corre-

lated to their primitive or secondary nature. Table 2 shows the richness of intratumoral vascularization prior to and after contrast agent administration on examinations done before RF. Levovist administration reduced the number of non-vascularized lesions on CD from 15/27 (55%) to 7/33 (21%). All vascularized lesions were metastases and so increased the number of vascularized lesions from 45% to 79%. Before Levovist injection only two lesions were massively vascularized (5–10 vessels); there were 15 after injection. When injected, Levovist started to amplify the Doppler signal with a mean delay of about 15 s (20–120 s) after injection with persistence of enhancement for approximately 8 min (3–15 min).

Table 2 Intratumoral vascularization evaluation on CD with/without DF before RF

Number of intratumor vessels	Without Levovist	After Levovist
0	15	7
1–4	10	11
5–10	2	15
Total	27[a]	33

[a]Missing data for six examinations.

Table 3 US mean size of lesions

	Time					
	D − 1	D + 1	2 months	4 months	6 months	1 year
US mean (mm)	29	37	38	36	39	36

At the 24 h examinations, the largest diameter of the lesions increased from 29 mm before RF ablation to 37 mm after (Table 3). In 18 lesions there was no detectable vascularization even with contrast agent (Fig. 1). Only four tumors presented an enhancement 24 h after RF ablation; two were incompletely sterilized large lesions of more than 55 mm; in the other two lesions, this transient enhancement, which was never found on subsequent examinations, disappeared 2 months later. At 2 months it was possible to compare CDUS and MRI results in 21 tumors. CDUS detected seven recurrent lesions while contrast-enhanced MRI showed recurrence in only five of them. The other two were subsequently confirmed as false positives of CDUS. Fourteen CDUS were negative, confirmed by MRI. At 4 months CDUS and MRI were compared in 20 lesions. CDUS was negative in 17 lesions, confirmed by MRI (Figs 2 and 3). Three CDUS were positive, confirmed only in two cases by MRI (Figs 4 and 5). For the third case, subsequent examinations demonstrated progressive tumoral evolution and so was a false negative of MRI (Fig. 6). At 6 and 12 months, 21 examinations (12 and 9, respectively) CDUS–MRI showed 12 confirmed true negatives of CDUS, six true positives and three false positives. In total, 62 CDUS performed between the 2nd and 12th months have been compared to MRI and to the clinical evolution of the treated lesions (Table 4).

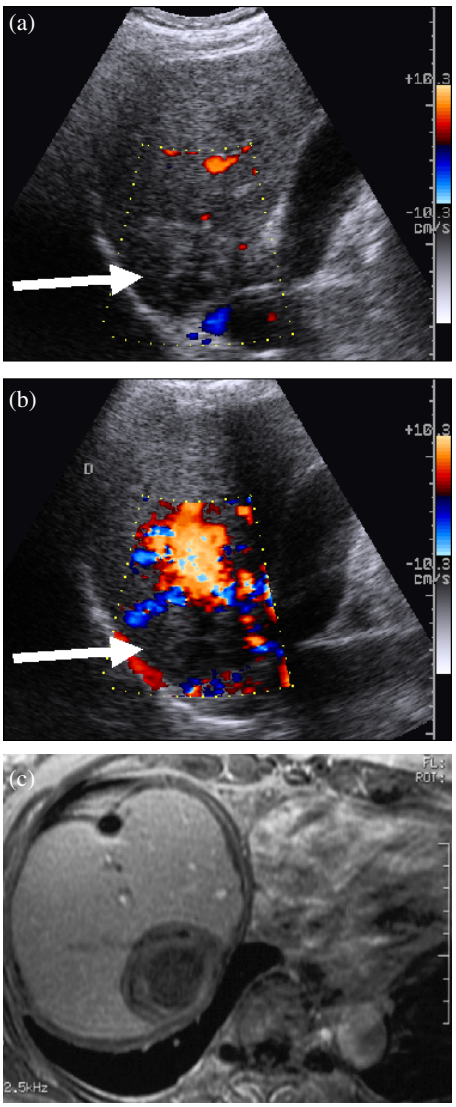

Figure 4 **Correlation between CDUS and MR at 4 months after RF ablation of HCC: (a, b) CDUS does not show any intratumoral vessel inside the treated area (arrow). (c) Contrast-enhanced T1-weighted MR confirms the complete sterilization of the treated lesion.**

Levovist was well tolerated; no secondary effect requiring emergency treatment arose (such as anaphylactic shock, cardiovascular troubles, discomfort, etc.) in 127 injections performed. One patient experienced a vague and ephemeral heating sensation and two others a cold sensation along the path of the injected vein, without consequence. For the two patients in whom an accidental extra-venous passage was encountered, there was no local pain, phlebitis or lymphangitis.

Discussion

MRI and to a lesser extent CT, are the preferred methods for the evaluation of the efficiency of RF ablation of

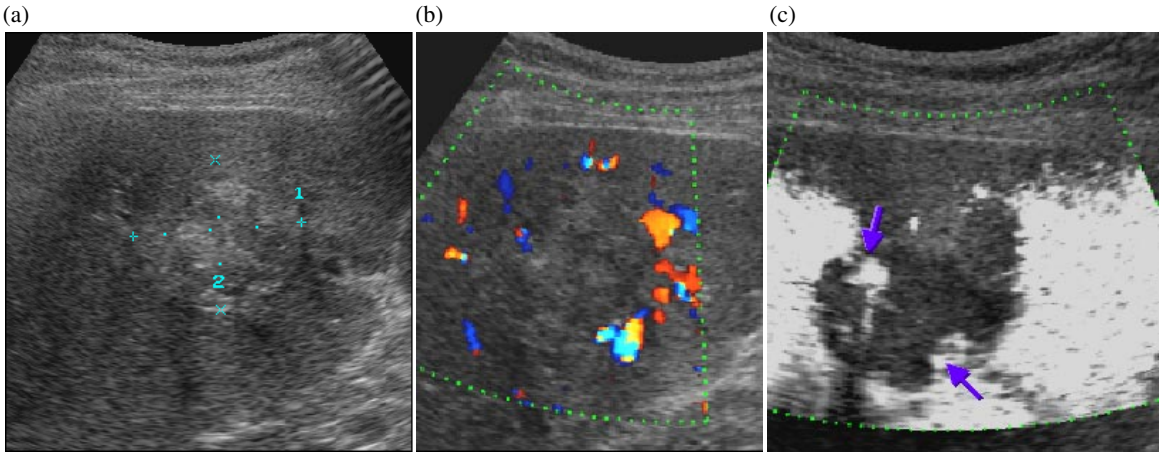

Figure 5 **Recurrence of colon tumor metastasis 6 months after RF ablation: (a) on mode B the lesion is hyperechoic and heterogeneous. (b) Color Doppler shows an intratumoral vessel suspicious of recurrence. (c) Contrast-enhanced dynamic flow US demonstrates at least two zones of intratumoral enhancement (arrows) that confirms the recurrence.**

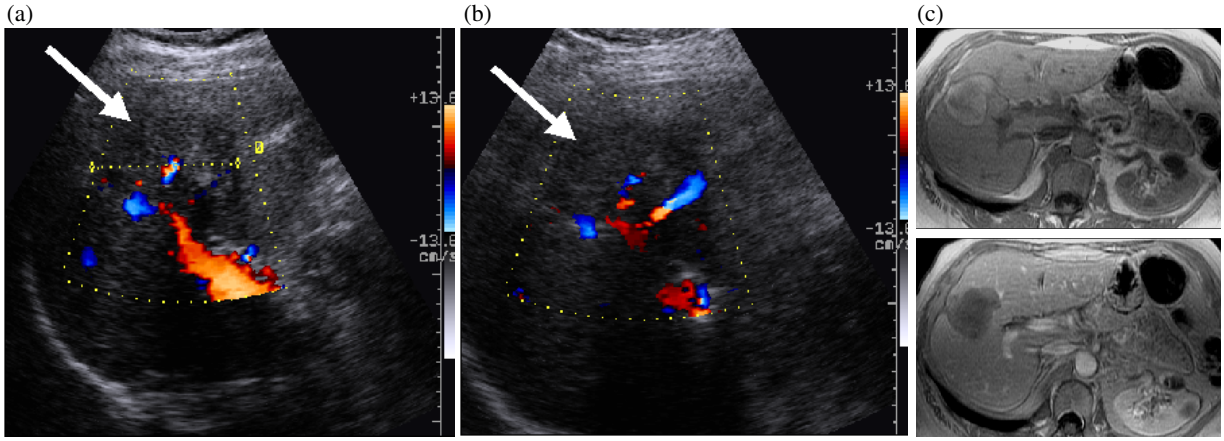

Figure 6 **A 61-year-old woman with metastasis of a colon tumor. Correlation between CDUS and MRI: (a, b) CDUS reveals vessels inside the treated lesion. (c) T1 weighted MRI scans before and after contrast enhancement show no intratumoral enhancement. Follow-up demonstrated that it was a recurrence and the patient underwent retreatment with RF ablation.**

primary and secondary hepatic tumors, and for the detection of local recurrences. There is a very good correlation between avascular non-enhanced necrotic zones visualized on MRI or CT and the size of coagulation necrosis measured by histological analysis[9]. Since 1999, contrast-enhanced ultrasonography has been used following RF to detect zones of hepatic recurrence[10]. This technique benefits from important technological improvements such as electronic processing of the ultrasound signal, as well as multiplication of the transducer's channels improving its resolution and the sensitivity of microvessel detection[11]. Imaging of slow flow has also been improved by innovations such as B flow Doppler mode (General Electric)[12] or dynamic flow (Toshiba)[13,14] among others. The use of a contrast agent in ultrasonography leads to signal enhancement of vessels and even the detection of neo-vessels as tiny as 40 μm in diameter[15].

After RF ablation, the size of the thermally induced lesion is much bigger than the non-treated tumor size. In our series, the mean diameter of lesions before and after RF increased from 29 to 37 mm. This shows that the gold standard of evaluation of therapeutic efficiency[16] based on tumor diameter measurements cannot be applied here. Evaluation must be based on the detection of residual vascularization or reappearance of vessels as an indication of recurrence.

New anti-cancer treatments (Melphalan, TNFα, Sugen, Neovastat) aim to block tumor development by preventing the formation of tumoral neo-vessels. Nowadays, the study of tumoral neovascularization represents an important objective for imaging research because of its

Table 4 Accuracy of CDUS for the detection of recurrence of hepatic lesions treated with RF ablation

CDUS (months)	TP	FP	FN	TN	Sensitivity	Specificity	PPV	NPV	No. of exams
2	5	2	0	14	1.00	0.87	0.71	1.00	21
4	3	0	0	17	1.00	1.00	1.00	1.00	20
6	3	2	0	7	1.00	0.78	0.60	1.00	12
12	3	1	0	5	1.00	0.83	0.75	1.00	9
Total	14	5	0	43	1.00	0.90	0.74	1.00	62

TP, true positive; FP, false positive; FN, false negative; TN, true negative; PPV, positive predictive value; NPV, negative predictive value.

role in the evaluation of the efficiency of these new anti-angiogenic treatments.

Hepatic tumors after treatment with RF ablation are often small, and in the majority of cases (56% in our series), non-enhanced color Doppler does not detect any intratumoral vessel before treatment, whereas vascularization was detected in about 70% of lesions when Levovist was administered. Levovist is thus of importance in increasing the number of lesions assessable by ultrasonography. For tumors in which vascularization cannot be assessed after Levovist injection before RF ablation, CDUS is of less value for immediate evaluation of treatment results. In addition to visualization of vessels through contrast media use, the ultrasound beam detects the explosion of injected microbubbles concentrated in the remaining vascularized tissue. This 'parenchymography' in theory allows the detection of viable tumor residue inside the thermally induced necrosis.

Liu *et al.*[17] showed that contrast-enhanced sonographic imaging was useful for detection of liver tumors and for guiding tumor ablation therapies as well as for evaluation of post-RF ablation efficiency. Solbiati *et al.*[10] evaluated contrast agent-enhanced color and power Doppler US in the detection of residual tumor after RF ablation of 20 colorectal liver metastases. Contrast-enhanced US performed 24 h after ablation demonstrated residual foci of enhancement in three tumors, whereas no US signals were seen in any tumor in non-enhanced studies. CT demonstrated small persistent foci of residual enhancement in these three tumors and in three additional lesions that were not seen at US (US sensitivity, 50%; specificity, 100%; diagnostic agreement with CT, 85%).

Bartolozzi *et al.*[18] investigated the ability of contrast agent-enhanced CDUS to evaluate the response of HCC to percutaneous ethanol injection (PEI). After PEI, blood flow signals were no longer detected in all the lesions that were found to be necrotic at spiral CT and biopsy (US specificity 100%). In 12 (92%) of 13 lesions containing residual viable tumor, intratumoral color signals corresponding to the enhancing areas at spiral CT were still identified at contrast-enhanced CDUS. They concluded that contrast-enhanced CDUS shows promise in assessing the therapeutic effect of PEI on HCC and in guiding additional treatment in cases of incomplete response.

The use by some investigators of new methods of Doppler (harmonic with pulse inversion) along with injection of contrast media in the evaluation of therapeutic response of hepatic tumors treated with RF ablation has given comparable results. Cioni *et al.*[19] in a series of 65 HCC treated with radiofrequency ablation could detect recurrences on harmonic power Doppler with Levovist with sensitivity of 90% compared to CT. On a series of 43 HCC, Meloni *et al.*[20] used the same sonographic technique and CT as a reference and they obtained a sensitivity of 83%.

With 100% sensitivity and 90% specificity, our technique using dynamic flow with Levovist compared to MRI achieves the same results overall, despite the fact that the majority of the cases were composed of secondary lesions generally less vascularized than HCC[21]. In the case of detection of suspect enhancement on CDUS, complementary MRI is advocated to confirm recurrence. In fact, between the 2nd and 12th months following treatment, MRI and the clinical evolution did not confirm recurrence except in 14 cases. Of four treated nodules enhanced on CDUS performed 24 h after RF ablation, two were in relation to residual foci of tumors confirmed by MRI and successfully retreated with RF; the other two showed a small peripheral enhancement and were false positives because the 2 months MRI and the subsequent CDUSs were perfectly normal without any enhancement. Thus, we concluded that these were early enhancement of post-operative hyperthermia and inflammation already described in many series[4,7], particularly because of their non-nodular appearance.

New agents such as SonoVue (BR1, Bracco, Italy), which is effective at low MI will shortly be available, which should increase the duration of enhancement, allow several passages and more effective parenchymography[22]. Furthermore, several research groups are developing new specific contrast media targeted on tumoral neovascularization.

The use of dynamic flow perfusion software is particularly interesting. Unlike standard methods of Doppler (color or power), the intravascular signal related to blood flow does not exceed the limits of the vessels, giving a more accurate imaging technique with less interobserver variability. Moreover, the appearance of new software to quantify the signal in an objective manner as a function of time would allow identification

of a particular kinetics of tumor tissue as distinct from inflammatory areas.

Conclusion

Our preliminary results show that contrast-enhanced CDUS is useful to confirm the absence of vascularity after treatment with RF ablation while the presence of enhancement must be confirmed by MRI. Further study will allow a better appreciation of its accuracy in long-term follow-up.

Acknowledgment

The authors are grateful to Lorna Saint Ange for editing.

References

[1] Solbiati L, Livraghi T, Goldberg SN *et al*. Percutaneous radio-frequency ablation of hepatic metastases from colorectal cancer: long-term results in 117 patients. Radiology 2001; 22: 159–66.

[2] McGhana JP, Dodd III GD. Radiofrequency ablation of the liver: current status. AJR Am J Roentgenol 2001; 176: 3–16.

[3] Morimoto M, Sugimori K, Shirato K *et al*. Treatment of hepatocellular carcinoma with radiofrequency ablation: radiologic–histologic correlation during follow-up periods. Hepatology 2002; 35: 1467–75.

[4] Goldberg SN, Gazelle GS, Mueller PR. Thermal ablation therapy for focal malignancy: a unified approach to underlying principles, techniques, and diagnostic imaging guidance. AJR Am J Roentgenol 2000; 174: 323–30.

[5] Dromain C, de Baere T, Elias D *et al*. Hepatic tumors treated with percutaneous radio-frequency ablation: CT and MR imaging follow-up. Radiology 2002; 223: 255–62.

[6] Chopra S, Dodd III GD, Chintapalli KN, Leyendecker JR, Karahan OI, Rhim H. Tumor recurrence after radiofrequency thermal ablation of hepatic tumors: spectrum of findings on dual-phase contrast-enhanced CT. AJR Am J Roentgenol 2001; 177: 381–7.

[7] Choi D, Lim HK, Kim SH *et al*. Hepatocellular carcinoma treated with percutaneous radio-frequency ablation: usefulness of power Doppler US with a microbubble contrast agent in evaluating therapeutic response-preliminary results. Radiology 2000; 217: 558–63.

[8] Ding H, Kudo M, Onda H *et al*. Evaluation of posttreatment response of hepatocellular carcinoma with contrast-enhanced coded phase-inversion harmonic US: comparison with dynamic CT. Radiology 2001; 221: 721–30.

[9] Goldberg SN, Gazelle GS, Compton CC, Mueller PR, Tanabe KK. Treatment of intrahepatic malignancy with radiofrequency ablation: radiologic–pathologic correlation. Cancer 2000; 88: 2452–63.

[10] Solbiati L, Goldberg SN, Ierace T, Dellanoce M, Livraghi T, Gazelle GS. Radio-frequency ablation of hepatic metastases: postprocedural assessment with a US microbubble contrast agent—early experience. Radiology 1999; 211: 643–9.

[11] Lassau N, Koscielny S, Avril MF *et al*. Prognostic value of angiogenesis evaluated with color Doppler high-frequency ultrasound for pre-operative assessment of melanomas. AJR Am J Roentgenol 2002; 178: 1547–51.

[12] Henri P, Tranquart F. B-flow ultrasonographic imaging of circulating blood. J Radiol 2000; 81: 465–7.

[13] Sato T. Technological description of dynamic flow. Toshiba Medical Review (Japanese edition) 2001; 80: 29–33.

[14] Koito K, Sato T. Contrast ultrasonography using dynamic flow—clinical usefulness in the evaluation of hepatobiliary and pancreatic diseases. Toshiba Medical Review (Japanese edition) 2001; 81: 30–8.

[15] Lassau N, Koscielny S, Opolon P *et al*. Evaluation of contrast-enhanced color Doppler ultrasound for the quantification of angiogenenis *in vivo*. Invest Radiol 2001; 36: 50–5.

[16] WHO. Handbook for reporting results of cancer treatment, Geneva, Switzerland: World Health Organization Offset Publication, 1979: 48.

[17] Liu JB, Goldberg BB, Merton DA, Rawool NM, Needleman L, Forsberg F. The role of contrast-enhanced sonography for radiofrequency ablation of liver tumors. J Ultrasound Med 2001; 20: 517–23.

[18] Bartolozzi C, Lencioni R, Ricci P, Paolicchi A, Rossi P, Passariello R. Hepatocellular carcinoma treatment with percutaneous ethanol injection: evaluation with contrast-enhanced color Doppler US. Radiology 1998; 209: 387–93.

[19] Cioni D, Lencioni R, Rossi S *et al*. Radiofrequency thermal ablation of hepatocellular carcinoma: using contrast-enhanced harmonic power Doppler sonography to assess treatment outcome. AJR Am J Roentgenol 2001; 177: 783–8.

[20] Meloni M, Goldberg SN, Livraghi T *et al*. Hepatocellular carcinoma treated with radiofrequency ablation: comparison of pulse inversion contrast-enhanced harmonic sonography, contrast-enhanced power Doppler sonography, and helical CT. AJR Am J Roentgenol 2001; 177: 375–80.

[21] Breedis C, Young G. The blood supply of neoplasms in the liver. Am J Pathol 1954; 30: 969–85.

[22] Bauer A, Solbiati L, Weissman N. Ultrasound imaging with SonoVue: low mechanical index real-time imaging. Acad Radiol 2002; 9 (Suppl 2): 282–4.