DOI:10.1093/jnci/dju239
First published online September 10, 2014

BRIEF COMMUNICATION

Randomized Trial of [131I] Metuximab in Treatment of Hepatocellular Carcinoma After Percutaneous Radiofrequency Ablation

Huijie Bian*, Jia-Sheng Zheng*, Gang Nan*, Rui Li, Changsheng Chen, Cai-Xia Hu, Yang Zhang, Bin Sun, Xi-Long Wang, Shi-Chang Cui, Jiao Wu, Jing Xu, Ding Wei, Xiaoyong Zhang, Haichun Liu, Wuwei Yang, Yong Ding, Jing Li, Zhi-Nan Chen

* Authors contributed equally to this work.

Manuscript received January 26, 2014; revised June 20, 2014; accepted July 1, 2014.

Correspondence to: Zhi-Nan Chen, PhD, Cell Engineering Research Center, Fourth Military Medical University, No. 169, Changle West Road, Xi'an 710032, China (e-mail: znchen@fmmu.edu.cn) or Jia-Sheng Zheng, Bachelor of Medicine, Oncology and Hepatobiliary Minimally Invasive Interventional Center, Beijing Youan Hospital, Capital Medical University, No. 8, Xi Tou Tiao, Youanmen wai, Fengtai District, Beijing 100069, China (e-mail: zhengjiasheng6@163.com).

To assess the efficacy of combining radioimmunoconjugate [131I] metuximab with radiofrequency ablation (RFA) in hepatocellular carcinoma (HCC) treatment compared with RFA alone, a single-center randomized controlled trial was conducted on 127 patients with Barcelona Clinic Liver Cancer staging system (BCLC) classifications of 0–B stage. Patients received either RFA followed by [131I] metuximab (n = 62) or RFA alone (n = 65). The primary outcome was overall tumor recurrence. Statistical tests were two-sided. The one- and two-year recurrence rates in the combination group were 31.8% and 58.5%, whereas those in the RFA group were 56.3% and 70.9%, respectively. The median time to overall tumor recurrence was 17 months in the combination group and 10 months in the RFA group (*P* = .03). The RFA-[131I] metuximab treatment showed a greater antirecurrence benefit than RFA in the metuximab target (ie, CD147)–positive subpopulation (*P* = .007). [131I] metuximab may yield prevention of tumor recurrence after RFA.

JNCI J Natl Cancer Inst (2014) 106(9): dju239 doi:10.1093/jnci/dju239

Radiofrequency ablation (RFA), as a minimally invasive treatment, is adopted for patients with early-stage hepatocellular carcinoma (HCC) who are not eligible for surgical resection or liver transplantation (1–3). It is also used for treating recurrent HCC after hepatectomy, ablation, or liver transplantation (4,5). However, one of major drawbacks is the high rate of disease recurrence with an adverse effect on patient survival (6).

Iodine [131I] metuximab injection (Licartin, Chengdu Huashen Biotechnology, Chengdu, China) is a radioimmunoconjugate generated by labeling metuximab directed against CD147 with iodine-131. CD147 is known as an extracellular matrix metalloproteinase inducer that was associated with hepatocarcinogenesis and tumor metastasis (7,8) and correlated with HCC grading (9). Combination of [131I] metuximab with liver transplantation or TACE (transcatheter arterial chemoembolization) showed an improved clinical efficacy in HCC therapy (10–12).

RFA causes cell damage by heat-induced coagulative necrosis and innate immunity activation. However, because of the limitations of heat dissipation and modest immune response, combining RFA with radioimmunotherapy probably has the potential to enhance outcome by directly targeted cell death (ie, hyperthermia, irradiation, and antibody) and indirectly adaptive immunity (13,14). We performed a randomized controlled trial (Chictr.org identifier: ChiCTR-TRC-10000837) to assess the efficacy and safety of [131I] metuximab combined with percutaneous RFA in patients with HCC compared with RFA alone.

The sample size was calculated using the historic cohort analysis of overall recurrence in this center. We needed 56 patients in each group (power of 90%, two-sided statistical significance level of 5%, 1:1 allocation) to detect a 30% recurrence rate of difference between groups. We also estimated and added 10% of patients who might be lost to follow-up. On the basis of these calculations, we estimated that we needed to enroll at least 124 patients.

Patients were recruited at a single center according to the guidelines from the European Association for the Study of the Liver (15): two imaging techniques or one imaging with α-fetoprotein greater than 400 ng/mL (n = 80) or cytological/histological evidence (n = 47). The eligibility criteria are described in the Supplementary Materials (available online). The study was conducted with the approval of the institutional ethics board of Beijing Youan Hospital of Capital Medical University. Written informed consent was obtained from each patient.

RFA was performed with computed tomography (CT) guidance. A single ablation with a cool-tip RFA single or cluster electrodes or multiple overlapping ablations with a cluster electrode were performed for tumors less than 3.0 cm and 3.0 cm and larger, respectively. If the ablation zone completely covered the tumor and ablation margin without new and residual lesions, the treatment was considered a complete destruction, otherwise an additional session of RFA was given. For patients with multiple and larger size tumors, two rounds of RFA were performed to ablate all lesions within 14 days depending on the liver function, but were considered one session. No more than three applications of RFA were given in a treatment. The injection of [131I] metuximab (27.75 MBq/kg) followed the last RFA within 30 days (median = 15 days; range = 3–29 days). Lugol's liquid was given starting three days before injection. A negative response to a subcutaneous metuximab injection was confirmed before the administration (16).

The primary outcome was overall recurrence. The criteria for establishing tumor recurrence is described in the Supplementary Materials (available online). The patients were followed up with once

every three months for the first year and once every six months thereafter. Tumor measurements were performed by blinded reviews. Secondary outcomes included overall survival and safety. Randomization was conducted using a computer program to achieve a balance between the two groups with stratification according to BCLC stage (stage 0–A vs stage B), tumor number (single vs multiple), and tumor size (<3 cm vs ≥3 cm). The data analysis was performed when overall 50% of the patients experienced a tumor recurrence. Outcomes were assessed according to the intention-to-treat principle. Comparisons between two groups were performed using the Student's *t* test for continuous data and the chi-square test for categorical data. Adverse events were compared with the Fisher's exact test. The proportional hazard assumption was checked by graphical inspection of the linearity of the hazards over time and log-log plots and by plotting Schoenfeld residuals over time. Statistical analyses were performed with SPSS 16.0 software (Statistical Product and Service Solutions, Chicago, IL).

As no competing risk events were observed, 1-Kaplan-Meier estimator was chosen to estimate the cumulative probability of recurrence, and two groups were compared using a log-rank test and confirmed by Cox proportional hazards models, stratified by BCLC stage, tumor number, and tumor size. All statistical tests were two-sided. A P value of less than .05 was considered statistically significant.

From April 13, 2010 to July 18, 2013, 127 patients were analyzed with 62 patients assigned to the RFA-[^{131}I] metuximab group and 65 patients assigned to the RFA group (Figure 1). There were no statistically significant differences between the two study groups in demographic characteristics (Table 1). The one- and two-year recurrence rates in the RFA-[^{131}I] metuximab group were 31.8% and 58.5%, whereas those in the RFA group were 56.3% and 70.9%, respectively. The median time to overall tumor recurrence was 17 months in the combination group and 10 months in the RFA group (HR = 0.60, 95% confidence interval [CI] = 0.38 to 0.96, P = .03) (Figure 2A). Using the log-rank test stratified by the three variables, the RFA-[^{131}I] metuximab group showed better results in terms of antirecurrence than the RFA group (Supplementary Table 1, available online). The prespecified subpopulation analysis showed an antirecurrence benefit for combination treatment over RFA in BCLC stage 0–A, tumor size 3 cm and larger, and single lesion (Supplementary Figure 1, available online). This suggests that combination therapy is beneficial to decreasing tumor recurrence in single larger tumors, possibly involving the adaptive immune response and the remodeling of the tumor microenvironment.

Biopsies from 47 patients were subjected to CD147 immunoreactive staining (Supplementary Figure 2, available online). The demographic characteristics and CD147 expression of this subpopulation are listed in Supplementary Table 2 (available online). Recurrence analysis shows the antirecurrence benefit of RFA-[^{131}I] metuximab treatment over RFA alone in the CD147-positive subpopulation (HR = 0.26, 95% CI = 0.09 to 0.79, P = .007) (Figure 2B). No statistically significant difference was detected in the CD147-negative subpopulation (HR = 0.41, 95% CI = 0.11 to 1.48, P = .12) (Figure 2C). At the cutoff date for analysis,

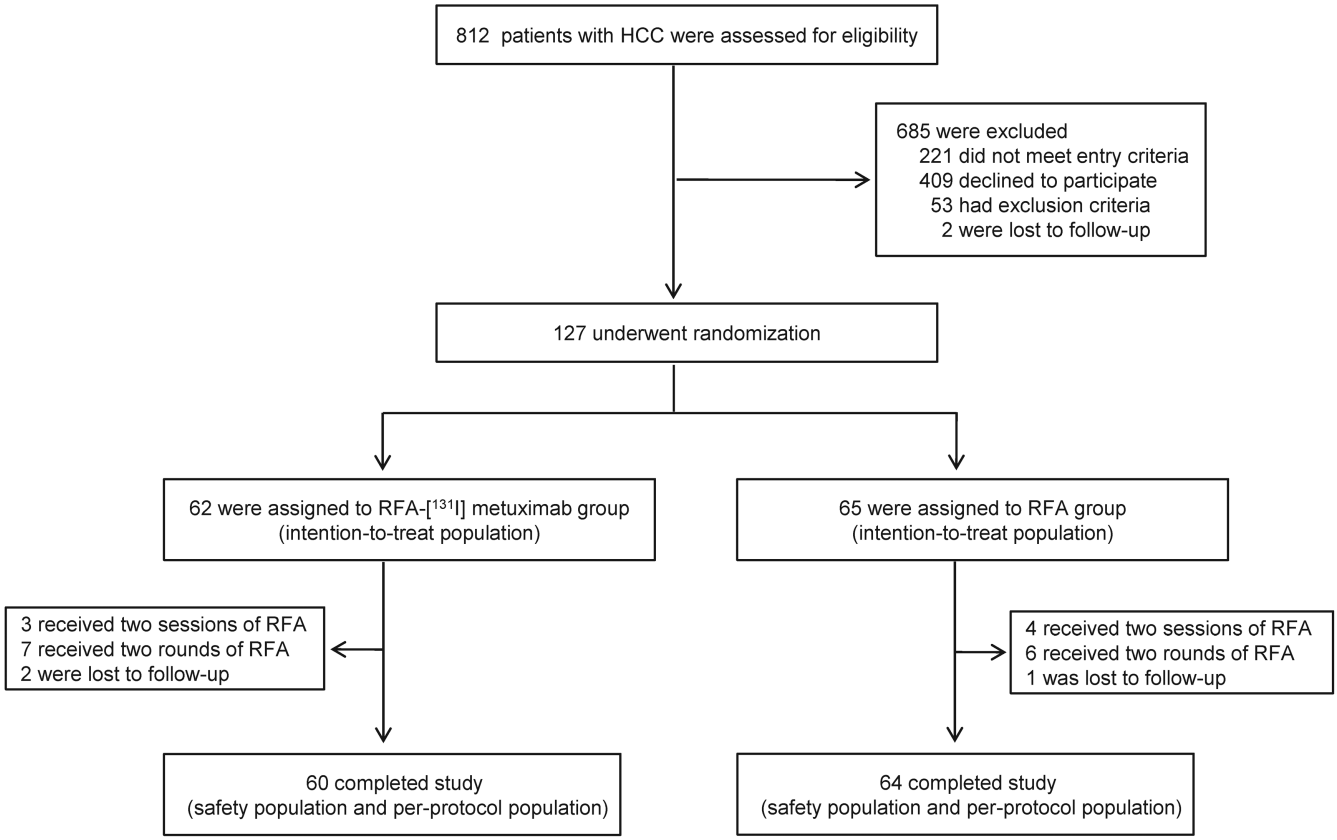

Figure 1. CONSORT diagram of the trial. HCC = hepatocellular carcinoma; RFA = radiofrequency ablation.

Table 1. Baseline patient characteristics

Characteristic	RFA-[^{131}I] metuximab (n = 62)	RFA (n = 65)	*P**
Age, y†			.26
Median	55	55	
Range	32 to 72	38 to 74	
Sex			.66
Male	55	56	
Female	7	9	
Cause of disease			.70
Hepatitis B only	54	53	
Hepatitis C only	6	8	
Alcohol only	0	1	
Unknown	2	3	
Hepatic cirrhosis			.95
Yes	59	62	
No	3	3	
Child-Pugh class			.07
A	56	51	
B	6	14	
BCLC stage			.72
0–A	45	49	
B	17	16	
Smoking			.79
Yes	30	33	
No	32	32	
AFP level, ng/mL			.17
<200	55	50	
≥200 and <400	3	9	
≥400	4	6	
Size of main tumor, cm†			.49
Mean	2.82	2.61	
SD	1.75	1.58	
Size range of tumor, cm			.98
<3	38	40	
≥3	24	25	
No. of tumors			.74
1	35	31	
2	11	14	
3	9	13	
4	7	7	
ALT, μ/L†			.99
Median	33.9	30.2	
Range	10.3–110.7	10.0–184.0	
AST, μ/L†			.27
Median	29.6	33.1	
Range	13.3–319.7	13.5–349.8	
TBIL, μmol/L†			.19
Median	18.2	19.7	
Range	5.2–43.6	5.7–58.6	
ALB, g/L†			.33
Median	37.7	36.3	
Range	28.7–49.3	27.5–48.9	
TT3, pmol/L†			.53
Median	1.9	2.0	
Range	1.2–2.4	1.1–3.0	
TT4, pmol/L†			.08
Median	93.1	109.5	
Range	62.3–118.9	62.8–174.4	
TSH, pmol/L†			.34
Median	3.8	2.9	
Range	0.3–10.5	0.8–8.8	

(Table continues)

Table 1 (Continued).

Characteristic	RFA-[131I] metuximab (n = 62)	RFA (n = 65)	P*
Previous therapy‡			
No	28	28	.81
Surgical resection	5	10	.20
Ablation	27	29	.90
TACE	23	28	.49

* *P* value was calculated using the chi-square test (two-sided). AFP = α-fetoprotein; ALB = albumin; ALT = alanine transaminase; AST = aspartate amino transferase; BCLC = Barcelona Clinic Liver Cancer staging system; HBV = hepatitis B virus; HCV = hepatitis C virus; RFA = radiofrequency ablation; SD = standard deviation; TBIL = total bilirubin; TT3 = total triiodotyronine; TT4 = total tetraiodothyronine; TACE = transcatheter arterial chemoembolization; TSH = thyroid stimulating hormone.

† *P* value was calculated using the Student's *t* test (two-sided).

‡ Patients might have received more than one type of therapy.

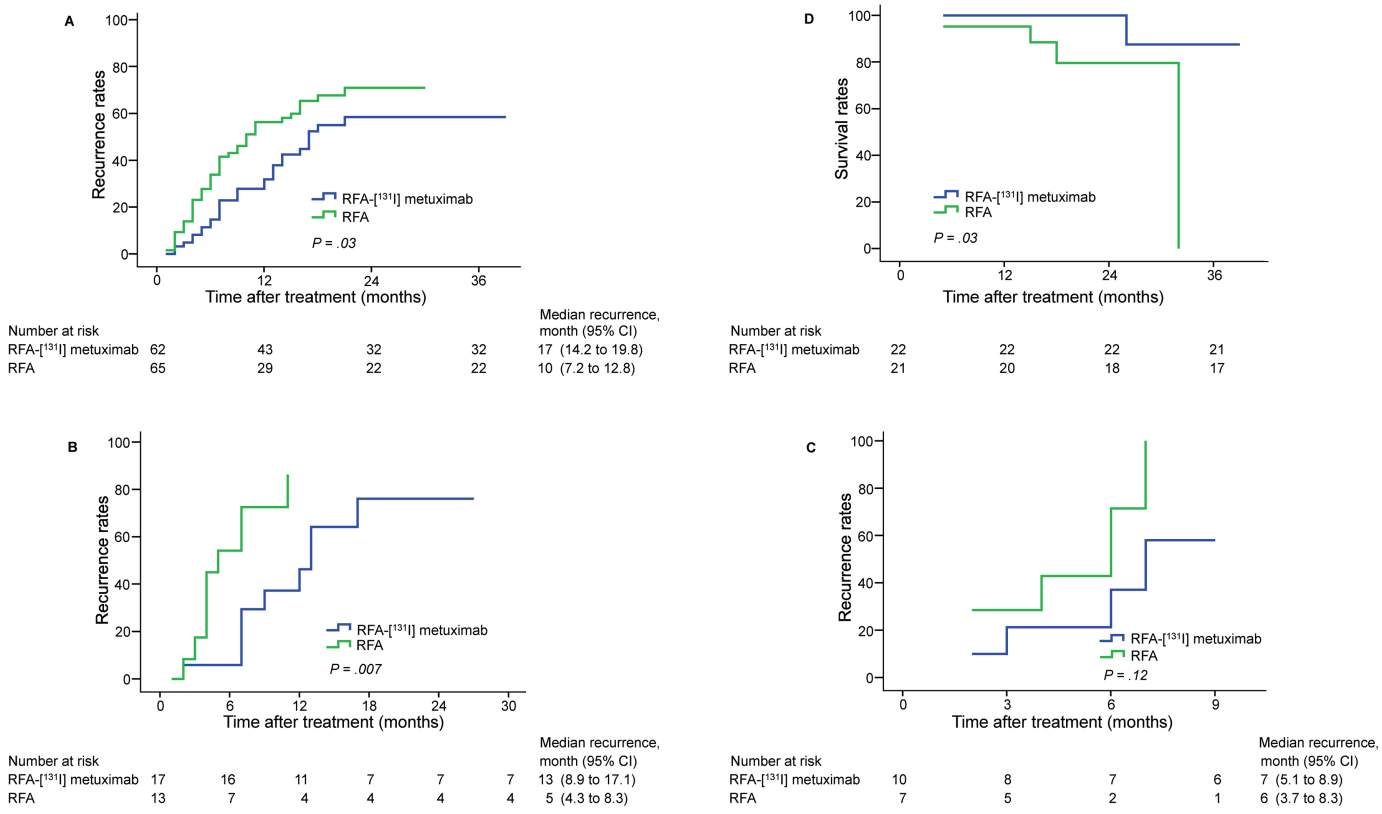

Figure 2. One minus Kaplan-Meier estimator of overall tumor recurrence and Kaplan-Meier analysis of overall survival for the RFA-[131I] metuximab and RFA groups. **A)** Overall tumor recurrence in the intention-to-treat population. **B)** Overall tumor recurrence in the CD147-positive subpopulation. **C)** Overall tumor recurrence in the CD147-negative subpopulation. **D)** Overall survival in the previously untreated subpopulation with BCLC stage 0–A. *P* values were calculated by log-rank test (two-sided). CI = confidence interval; RFA = radiofrequency ablation.

11 patients in the RFA-[131I] metuximab group and 15 patients in the RFA group had died of tumor recurrence. The one- and two-year overall survivals for the RFA-[131I] metuximab group were 93.5% and 84.7%, and those for the RFA group were 90.1% and 76.4%, respectively (HR = 0.66, 95% CI = 0.30 to 1.46, *P* = .30). In previously untreated tumors with BCLC stage 0–A subgroup (n = 43), the RFA-[131I] metuximab treatment (n = 22) showed better overall survival than the RFA treatment (n = 21) (HR = 0.11, 95% CI = 0.01 to 1.10, *P* = .03) (Figure 2D). Adverse events were predominantly grades 1 or 2 in clinical and laboratory toxicities (Supplementary Table 3, available online). Thyroid function in the RFA-[131I] metuximab group was not altered by [131I] metuximab treatment (Supplementary Table 4, available online). The HBV replication was suppressed minor by [131I] metuximab treatment (*P* = .049) (Supplementary Table 5, available online). No serious adverse events or treatment-related deaths were observed.

Limitations of the study were single center, small sample size, mixed population of previously treated and untreated individuals, shorter follow-up, limited number of biopsies, and single use of [131I] metuximab.

A future study will be designed to overcome the limitations and confirm our findings.

In conclusion, this study demonstrated a beneficial treatment effect of [^{131}I] metuximab after radiofrequency ablation on prevention of tumor recurrence in patients with HCC, a CD147-targeted therapeutic strategy for tumor recurrence control, and a prolonging of overall survival of early-stage, untreated tumors.

References

1. Omata M, Lesmana LA, Tateishi R, et al. Asian Pacific Association for the Study of the Liver consensus recommendations on hepatocellular carcinoma. *Hepatol Int*. 2010;4(2):439–474.
2. Bruix J, Sherman M. Management of hepatocellular carcinoma: an update. *Hepatology*. 2011;53(3):1020–1022.
3. European Association For The Study Of The Liver, European Organisation For Research And Treatment Of Cancer. EASL-EORTC clinical practice guidelines: management of hepatocellular carcinoma. *J Hepatol*. 2012;56(4):908–943.
4. Peng ZW, Zhang YJ, Liang HH, et al. Recurrent hepatocellular carcinoma treated with sequential transcatheter arterial chemoembolization and RF ablation versus RF ablation alone: a prospective randomized trial. *Radiology*. 2012;262(2):689–700.
5. Ho CK, Chapman WC, Brown DB. Radiofrequency ablation of recurrent hepatocellular carcinoma in a patient after liver transplantation: two-year follow-up. *J Vasc Interv Radiol*. 2007;18(11):1451–1453.
6. Tiong L, Maddern GJ. Systematic review and meta-analysis of survival and disease recurrence after radiofrequency ablation for hepatocellular carcinoma. *Br J Surg*. 2011;98(9):1210–1224.
7. Wu J, Ru NY, Zhang Y, et al. HAb18G/CD147 promotes epithelial-mesenchymal transition through TGF-β signaling and is transcriptionally regulated by Slug. *Oncogene*. 2011;30(43):4410–4427.
8. Zhao P, Zhang W, Wang SJ, et al. HAb18G/CD147 promotes cell motility by regulating annexin II-activated RhoA and Rac1 signaling pathways in hepatocellular carcinoma cells. *Hepatology*. 2011;54(6):2012–2024.
9. Zhang Q, Zhou J, Ku XM, et al. Expression of CD147 as a significantly unfavorable prognostic factor in hepatocellular carcinoma. *Eur J Cancer Prev*. 2007;16(3):196–202.
10. Xu J, Shen ZY, Chen XG, et al. A randomized controlled trial of Licartin for preventing hepatoma recurrence after liver transplantation. *Hepatology*. 2007;45(2):269–276.
11. Wu L, Yang YF, Ge NJ, et al. Hepatic artery injection of ^{131}I-labelled metuximab combined with chemoembolization for intermediate hepatocellular carcinoma: a prospective nonrandomized study. *Eur J Nucl Med Mol Imaging*. 2012;39(8):1306–1315.
12. Wu L, Yang YF, Ge NJ, et al. Hepatic arterial iodine-131-labeled metuximab injection combined with chemoembolization for unresectable hepatocellular carcinoma: interim safety and survival data from 110 patients. *Cancer Biother Radiopharm*. 2010;25(6):657–663.
13. Chu KF, Dupuy DE. Thermal ablation of tumours: biological mechanisms and advances in therapy. *Nat Rev Cancer*. 2014;14(3):199–208.
14. Greten TF, Korangy F. Radiofrequency ablation for the treatment of HCC--maybe much more than simple tumor destruction? *J Hepatol*. 2010;53(4):775–776.
15. Bruix J, Sherman M, Llovet JM, et al. Clinical management of hepatocellular carcinoma. Conclusions of the Barcelona-2000 EASL conference. European Association for the Study of the Liver. *J Hepatol*. 2001;35(3):421–430.
16. Chen ZN, Mi L, Xu J, et al. Targeting radioimmunotherapy of hepatocellular carcinoma with iodine (131I) metuximab injection: clinical phase I/II trials. *Int J Radiat Oncol Biol Phys*. 2006;65(2):435–444.

Funding

This work was supported by grants from the National Science and Technology Major Project (2012ZX10002-015 and 2013ZX09301301), the National Natural Science Foundation of China (81172144), and the Hi-tech Research and Development Program of China (2012AA020806).

Notes

The principal investigators (ZN Chen and JS Zheng) had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. All authors reviewed the manuscript draft and approved the final version for submission. No authors reported financial disclosures. The sponsors had no role in the design, conduct, or interpretation of the study.

Affiliations of authors: Cell Engineering Research Center and Department of Cell Biology, State Key Laboratory of Cancer Biology (HB, GN, YZ, XLW, JW, JX, DW, ZNC) and Department of Health Statistics (CC), Fourth Military Medical University, Xi'an, China; Oncology and Hepatobiliary Minimally Invasive Interventional Center, Beijing Youan Hospital, Capital Medical University, Beijing, China (JSZ, RL, CXH, BS, SCC); Radionuclide Diagnosis and Treatment Center, No. 401 Hospital of China Nuclear Industry, Beijing, China (XZ, HL); Tumor Minimally Invasive Therapy Division (WY, JL) and Department of Nuclear Medicine (YD), Hospital 307 of PLA, Beijing, China.