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Cryoprotective therapy for hepatocellular carcinoma: Study of 51 patients with a single lesion ☆

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ABSTRACT

Percutaneous cryoablation is a potentially curative treatment for hepatocellular carcinoma (HCC). After liver cryosurgery, rapid elevations of transaminases and bilirubin are common, but are usually transient and normalize within a few days. This study retrospectively reviewed clinical data from 51 patients who underwent liver cryoablation in our hospital during the past 4.5 years. Sixty-six percutaneous cryoablations were performed in these patients and transaminase and bilirubin levels before and after the procedure were observed. Although most patients received liver-protective treatment before cryosurgery, transaminase levels were double (mean alanine transaminase (ALT) and aspartate transaminase (AST) were 71 U/L and 85 U/L, respectively) the normal ranges in our hospital. One day after cryosurgery, ALT and AST had increased 3.3-fold (peak mean was 241 U/L) and 5-fold (peak mean was 427 U/L), respectively, but were close to the preoperative level 5 days post-cryosurgery. No significant increase of serum bilirubin was observed. Serum transaminase and bilirubin levels were compared between hepatitis B positive and hepatitis B negative patients. Only in the hepatitis B positive group were total bilirubin (74 μmol/L/23 μmol/L = 3.2) and direct bilirubin (45 μmol/L/12 μmol/L = 3.8) more than 3 times the preoperative level 7–9 days after treatment. Overall, ALT and AST are valuable as indicators of liver function impairment following cryosurgery. In patients with hepatitis B virus, serum bilirubin was 3 times the preoperative level 7–9 days after cryosurgery. Liver-protective treatment may alleviate liver function impairment due to cryosurgery.

Introduction

Hepatocellular carcinoma (HCC) is one of the most common malignancies worldwide and its incidence is increasing [4]. In about 80% of patients, HCC is associated with chronic liver disease (i.e., hepatitis, cirrhosis), which has major implications for the prognosis and therapeutic options [9]. Many patients are unsuitable for tumor resection because of factors such as poor hepatic reserve, cirrhosis or the presence of multicentric tumors or extrahepatic disease [2,8]. Fortunately, non-surgical percutaneous tumor ablation therapies – namely, percutaneous ethanol injection, microwave coagulation, radiofrequency ablation and cryoablation – have been introduced and now play an important role in the treatment of HCC [16,22]. Cryoablation is an attractive option because the therapy can prolong the patient's life and the complications are acceptable. A long-term study of medium to large lesions (mean diameter 4.6 cm) treated with cryoablation and/or transarterial chemoembolization showed a 5-year survival rate of 23% and a local progression rate of 24% [14,23]. The complication rate following cryoablation may be as high as 40%; side effects include liver parenchyma fracture, cryoshock, biliary fistula, hemorrhage, cold-induced lesions in nearby structures, thrombocytopenia and coagulopathy, but these can be reduced by skilled operators, strict patient selection and symptomatic treatment [15,18,19,21]. Ablating the tumor as completely as possible

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inevitably damages the normal liver tissue, affecting liver function. Preservation of liver function is at least as important as ablation of HCC nodules in patients with already impaired liver function [10]. After liver cryosurgery, rapid elevation of transaminases [6,7,17,20,26] and bilirubin [1,7,11] is common, but the elevation is usually transient and commonly normalizes within a few days.

To gather detailed information on the effect of cryoablation on liver function, 51 HCC patients were observed for hepatic functional reserve after 66 percutaneous cryoablations. Liver-protective therapy and symptomatic treatment were provided to these patients and serum transaminase and bilirubin levels were compared between hepatitis B virus positive and hepatitis B virus negative patients.

Materials and methods

Ethics

The study protocol received ethical approval from the Regional Ethics Committee of Guangzhou Fuda Cancer Hospital. Written informed consent was obtained from each participant in accordance with the Declaration of Helsinki.

Patient selection

In all patients, the diagnosis of unresectable HCC was confirmed by liver pathology, imaging and tumor markers. Ideal patients for this research were those with: only one significant tumor in the liver; Karnofsky performance status score $\geqslant$70; platelet count $\geqslant 100 \times 10^9$/L; white blood cell count $\geqslant 3 \times 10^9$/L; neutrophil count $\geqslant 2 \times 10^9$/L; hemoglobin $\geqslant$ 90 g/L; prothrombin time international normalized ratio $\geqslant$ 1.5; hepatic tumor not obviously invading the gallbladder, diaphragm or large vessels; absence of level 3 hypertension, severe coronary disease, myelosuppression, respiratory disease and acute or chronic infection; and adequate hepatic function (total bilirubin (T.BIL) < 75 μmol/L, direct bilirubin (D.BIL) < 39 μmol/L and Child–Pugh score A or B) and renal function (serum creatinine < 130 μM, serum urea < 10 mM). Between January, 2005 and January, 2008, 38 patients met our inclusion criteria and received simple hepatic cryoablation; from January, 2008 to July, 2012, 51 patients were enrolled in the study and received cryoprotective therapy.

Cryotherapy

Each procedure comprised two freeze–thaw cycles accomplished using an argon gas-based cryosurgical unit (Endocare, Irvine, CA, USA). Cryoprobes (3 mm in diameter) were inserted into the center of the tumor mass under ultrasonographic guidance, each reaching a temperature of −150 °C at the tip of the probe. The duration of freezing was dependent on the achievement of an ice ball that extended 1 cm beyond the boundary of the tumor and was visible as a hypoechoic region on ultrasonography. Generally, the maximal freezing time was 15 min, followed by thawing for 5 min; this cycle was then repeated. For masses <6 cm in long diameter, two or three cryoprobes were placed within the center of the tumor, to ensure freezing of the entire mass; for masses with a long diameter of 6–10 cm, the tumor was divided into two parts that were treated in turn, usually at an interval of 1 week; for masses with a long diameter of 10–18 cm, the tumor was divided into three parts treated at intervals of usually 1 week. The tracts formed were sealed with fibrin glue immediately after removal of the cryoprobes to ensure hemostasis.

Tests of hepatic functional reserve

Hepatic function was measured using an automatic biochemical analyzer (Hitachi-7100; Hitachi, Tokyo, Japan); alanine transaminase (ALT) and aspartate transaminase (AST) were determined with a specialized reagent kit using the velocity method and T.BIL and D.BIL were determined with a kit using the vanadate method (Biosino Biotechnology and Science Inc., Beijing, China).

Blood was drawn in the morning on an empty stomach. The tests were performed every 1–3 days until patient discharge. Normal ranges are 5–35 U/L for ALT, 8–40 U/L for AST, 0–25.5 μmol/L for T.BIL and 0–13 μmol/L for D.BIL. Values higher than the upper limits were considered abnormal.

Liver-protective therapy and symptomatic treatment

Diammonium glycyrrhizinate capsules (150 mg twice per day orally; Chia Tai Tianqing Pharmaceutical Co. Ltd, Jiangsu, China) [25] and Atomolam (reduced glutathione tablets, 180 mg/day intravenous injection; Yaopharma, Chongqing, China) [24] were administered to all patients. In patients with abnormal transaminase, the two drugs were given until the liver function returned to normal; in patients with normal transaminase, the drugs were usually ceased 3–4 days post-cryoablation to help maintain normal liver function.

In patients with thrombocytopenia (platelet count $\leqslant 150 \times 10^9$/L), platelet count was closely supervised and recombinant human interleukin-11 (rhIL-11, 25 μg/kg/day; Northland Biotech, Beijing, China) was given if coagulopathy (platelet count $\leqslant 100 \times 10^9$/L) was detected [21]. For treatment of liver hemorrhage after cryosurgery, regular hemostatic agents were used; for treatment of liver abscess, antibiotic and drainage treatment was instituted.

Evaluation and statistical analysis

All cases of impaired hepatic function in this study were unambiguous. Complications were recorded and classified in accordance with the Common Terminology Criteria of Adverse Events v4.0. Radiographic local tumor control was assessed using image-guided tumor ablation criteria [5]. Abdominal ultrasonography was performed both 1 day and 1 week after treatment. The revised Response Evaluation Criteria in Solid Tumors v1.1 were used to assess the response of the hepatic tumors [3]. Bonferroni's multiple comparison tests were used for comparisons between days. Test results were expressed as the mean ± standard error and statistical significance was indicated by $P < 0.05$, $P < 0.01$ or $P < 0.001$. All analyses were conducted using GraphPad software (San Diego, CA, USA).

Results

Clinical data

Detailed data for 38 patients before simple hepatic cryoablation (51 sessions) are recorded in Table 1. Tumor long diameters were <6 cm in 28 patients, 6–10 cm in 8 and >10 cm in two. Twenty-two (58%) had stage IV disease, 12 (32%) had a history of hepatitis, 174 (45%) had initially been treated surgically and 11 (29%) had been treated with systemic chemotherapy in other centers, and 16 (43%) had thrombocytopenia. Detailed data for 51 patients before cryoprotective therapy (66 sessions) are recorded in Table 2. Tumor long diameters were <6 cm in 38 patients, 6–10 cm in 11 and >10 cm in two. Thirty-one (61%) had stage IV disease, 14 (27%) had a history of hepatitis, 24 (47%) had initially been treated surgically and 15 (29%) had been treated with systemic chemotherapy in other centers, and 24 (47%) had thrombocytopenia.

Table 1
Detailed data of 38 patients before simple hepatic cryoablation.

No.	Gender/age	Nationality	Cancer stage	ALT/AST (U/L)	T/D.BIL (μmol/L)	Hepatitis	No.	Gender/age	Nationality	Cancer stage	ALT/AST (U/L)	T/D.BIL (μmol/L)	Hepatitis	No.	Gender/age	Nationality	Cancer stage	ALT/AST (U/L)	T/D.BIL(μmol/L)	Hepatitis
1	M/79	China	IV	42/79	11/6	–	17	M/41	China	IV	26/53	26/14	–	26	M/64	China	III	15/52	20/8	–
2	M/42	China	IV	35/82	10/5	B+					119/188	23/11						11/39	11/5	
3	M/41	China	IV	114/106	41/20	–	18	M/30	China	III	208/141	36/22	–	27	M/40	Indonesia	IV	50/53	15/6	–
4	M/56	China	II	83/78	50/30	–	19	M/63	China	IV	221/186	27/15	B+					48/48	9/3	
5	F/53	China	III	18/121	33/12	–	20	M/55	Philippines	II	128/55	30/11	B+	28	M/69	Indonesia	III	18/34	12/3	–
6	F/56	China	II	77/43	20/8	B+					114/85	21/11						19/20	25/14	
7	F/57	China	II	36/31	21/10	B+					36/51	12/6		29	F/50	Vietnam	IV	17/24	7/1	–
8	M/49	Malaysia	IV	90/73	40/16	–	21	M/49	China	IV	74/114	39/28	B+	30	M/66	Indonesia	III	76/168	34/16	B+
9	M/45	China	IV	36/40	74/36	–	22	M/38	China	IV	92/43	28/11	–	31	M/41	China	IV	47/76	24/12	B+
10	M/76	Indonesia	II	11/20	18/7	C+					21/35	13/7		32	M/59	China	II	50/52	10/3	C+
11	M/45	China	IV	238/151	8/2	–	23	M/54	China	III	75/82	9/5	B+	33	M/30	Indonesia	IV	55/213	26/17	–
12	M/44	China	II	18/22	10/3	–					98/60	19/6		34	M/41	China	IV	24/60	27/11	–
13	M/40	China	IV	16/55	12/9	B+					69/87	9/2						110/137	25/10	
				25/37	20/10		24	M/56	China	III	21/189	26/14	–	35	F/96	Philippines	IV	97/100	22/11	–
14	M/56	China	IV	72/69	55/24	–					28/95	20/8		36	M/69	China	IV	54/197	26/15	–
15	M/81	China	IV	30/35	13/7	–	25	M/36	China	IV	11/39	11/5	–	37	F/80	Thailand	II	25/25	15/6	–
16	M/48	China	III	43/126	23/11	–					15/52	18/9		38	M/40	China	IV	65/92	14/6	–

Note: M, male; F, female; ALT, alanine aminotransferase; AST, aspartate aminotransferase; TBIL, total bilirubin; DBIL, direct bilirubin.

Table 2
Detailed data of 51 patients before cryoprotective therapy.

No.	Gender/ age	Nationality	Cancer stage	ALT/ AST (U/L)	T/D.BIL (μmol/ L)	Hepatitis	No.	Gender/ age	Nationality	Cancer stage	ALT/ AST (U/L)	T/ D.BIL(μmol/ L)	Hepatitis	No.	Gender/ age	Nationality	Cancer stage	ALT/ AST (U/L)	T/D.BIL (μmol/ L)	Hepatitis
1	M/79	China	IV	85/78	11/6	–	22	M/41	China	IV	26/53	18/9	–	35	M/30	China	III	15/52	20/8	–
2	M/42	China	IV	35/82	10/5	–					119/ 188	23/11						11/39	11/5	
3	M/41	China	IV	114/ 106	41/20	–	23	M/56	China	IV	508/ 231	36/22	–	36	M/40	Indonesia	IV	24/60	15/6	–
4	M/56	China	II	42/69	50/30	–	24	M/63	China	IV	26/51	27/15	B+					50/53	9/3	
5	M/58	China	III	41/40	17/9	–	25	M/50	China	IV	231/ 160	25/16	B+/	37	M/65	China	III	61/74	7/3	–
6	F/83	China	III	18/ 121	33/12	B+	26	M/55	China	II	128/ 55	21/11	B+	38	M/69	Indonesia	III	18/34	12/3	–
7	F/49	China	II	77/43	20/8	B+					314/ 115	12/6						19/20	10/3	
8	F/57	China	II	36/31	20/10	B+					221/ 186	30/11		39	F/50	Vietnam	IV	17/24	7/1	–
9	M/56	Malaysia	IV	72/69	40/16	–	27	M/38	China	IV	74/ 134	39/28	B+	40	M/66	Indonesia	III	76/ 168	34/16	B+
10	M/54	China	II	70/65	16/6	–	28	M/57	Indonesia	IV	28/38	20/13	–	41	M/53	China	IV	49/62	22/11	–
11	M/62	China	III	131/ 271	26/18	B+					31/33	36/26		42	M/47	Philippines	IV	15/27	18/7	–
12	M/45	China	IV	75/ 118	25/18	–	29	M/69	China	IV	67/55	34/13	–					63/24	43/22	
13	M/61	China	IV	28/ 121	63/42	–	30	M/38	China	IV	50/ 114	24/14	–	43	M/57	China	III	94/ 103	24/16	–
14	M/45	China	IV	36/40	74/36	–	31	M/49	China	III	92/43	28/11	–	44	M/41	China	IV	47/76	24/12	B+
15	M/76	China	II	11/20	18/7	–					21/35	13/7		45	M/59	China	II	50/52	22/11	B+
16	M/45	China	IV	238/ 151	8/2	–	32	M/54	Vietnam	IV	75/82	9/5	B+	46	M/64	Indonesia	IV	65/92	26/17	–
17	M/44	China	II	18/22	10/3	–					98/50	19/6		47	M/41	China	IV	48/48	27/11	–
18	M/40	Indonesia	IV	16/55	12/9	B+					69/87	9/2						110/ 137	25/10	
				15/37	23/11		33	M/30	China	III	21/ 189	26/14	–	48	F/96	Philippines	IV	97/ 100	25/14	C+/
19	M/56	China	IV	90/73	55/24	–					15/52	20/8		49	M/69	China	IV	54/ 197	26/15	–
20	M/53	China	IV	30/35	13/7	–	34	M/36	China	III	11/39	11/5	–	50	F/80	Thailand	IV	25/25	15/6	–
21	M/48	China	III	4/126	21/10	–					21/ 189	26/14		51	M/40	China	IV	55/ 213	14/6	–

Note: M, male; F, female; ALT, alanine aminotransferase; AST, aspartate aminotransferase; TBIL, total bilirubin; DBIL, direct bilirubin.

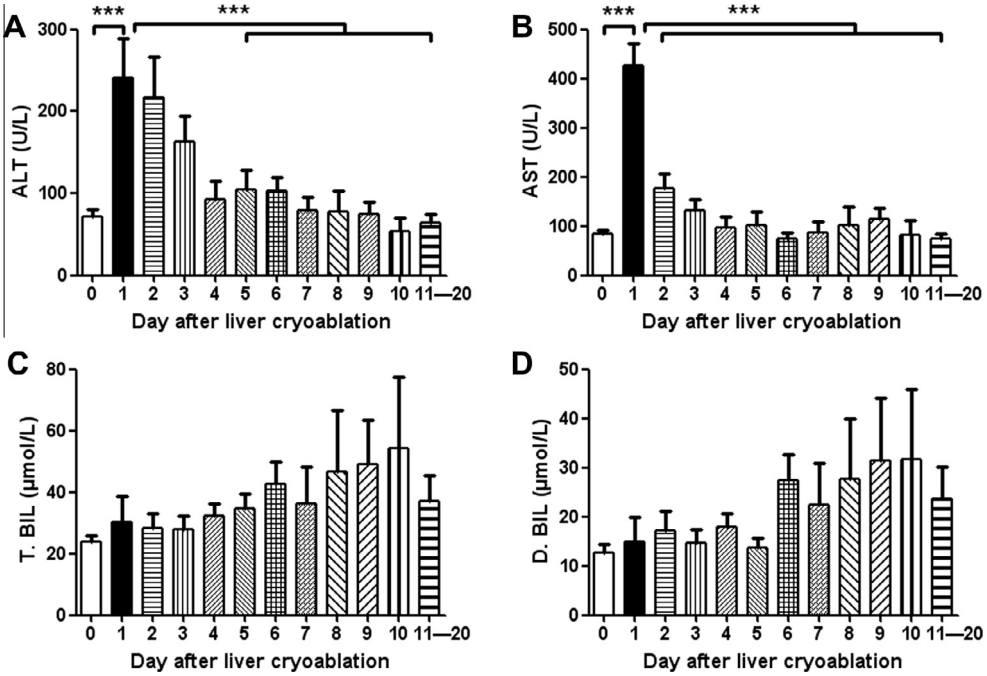

Fig. 1. Changes in hepatic functional reserve after 66 sessions of liver cryosurgery. Bonferroni's multiple comparison test was used to compare different days. The number of test results obtained varied between days (66, 18, 18, 16, 10, 15, 8, 9, 6, 8, 4 and 22 results were obtained on day 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10 and 11–20, respectively); thus, days with fewer than three test results were merged with neighboring days. Indices of hepatic functional reserve were alanine transaminase (ALT) (A), aspartate transaminase (AST) (B), total bilirubin (T.BIL) (C) and direct bilirubin (D.BIL) (D). $^{***}P < 0.001$.

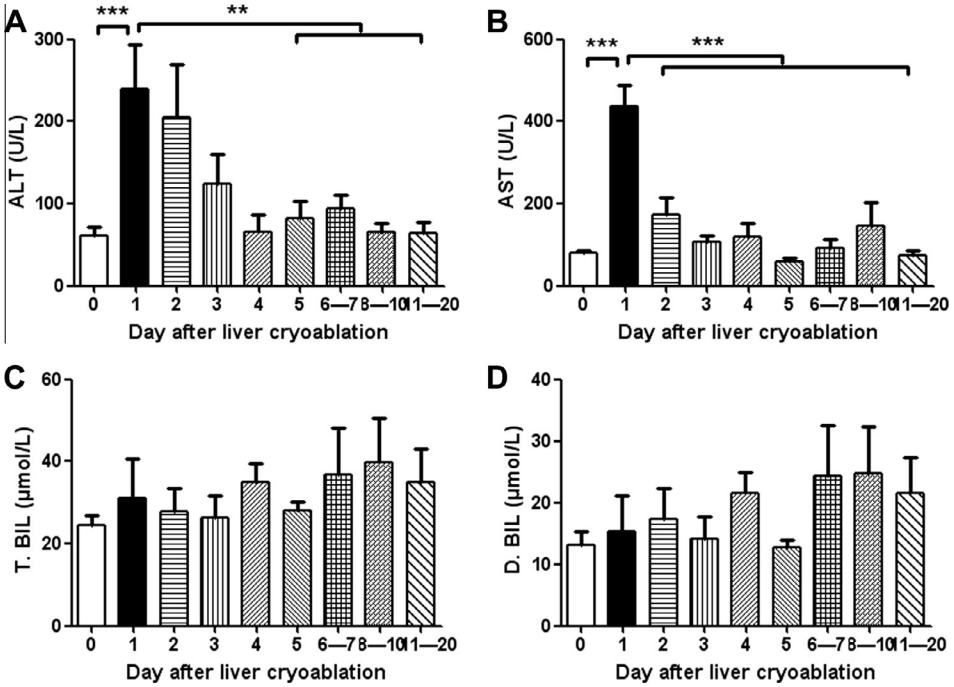

Fig. 2. Changes in the hepatic functional reserve of 37 non-hepatitis patients. Bonferroni's multiple comparison test was used to compare different days. The number of test results obtained varied between days (48, 16, 12, 10, 6, 11, 10, 12 and 18 results were obtained on day 0, 1, 2, 3, 4, 5, 6–7, 8–10 and 11–20, respectively); thus, days with fewer than three test results were merged with neighboring days. Indices of hepatic functional reserve were alanine transaminase (ALT) (A), aspartate transaminase (AST) (B), total bilirubin (T.BIL) (C) and direct bilirubin (D.BIL) (D). $^{**}P < 0.01$, $^{***}P < 0.001$.

Perioperative outcomes

All percutaneous cryoablations of hepatic lesions were performed successfully. No severe complications (such as liver rupture or failure, myoglobinuria or acute renal failure) were discovered post-cryoablation. Many mild side effects occurred after cryosurgery, but the affected patients recovered with or without symptomatic treatment. Slight liver hemorrhage occurred after 27 sessions (23%), but all cases resolved within 5 days with injection of hemostatic agents. Liver capsular rupture occurred during six

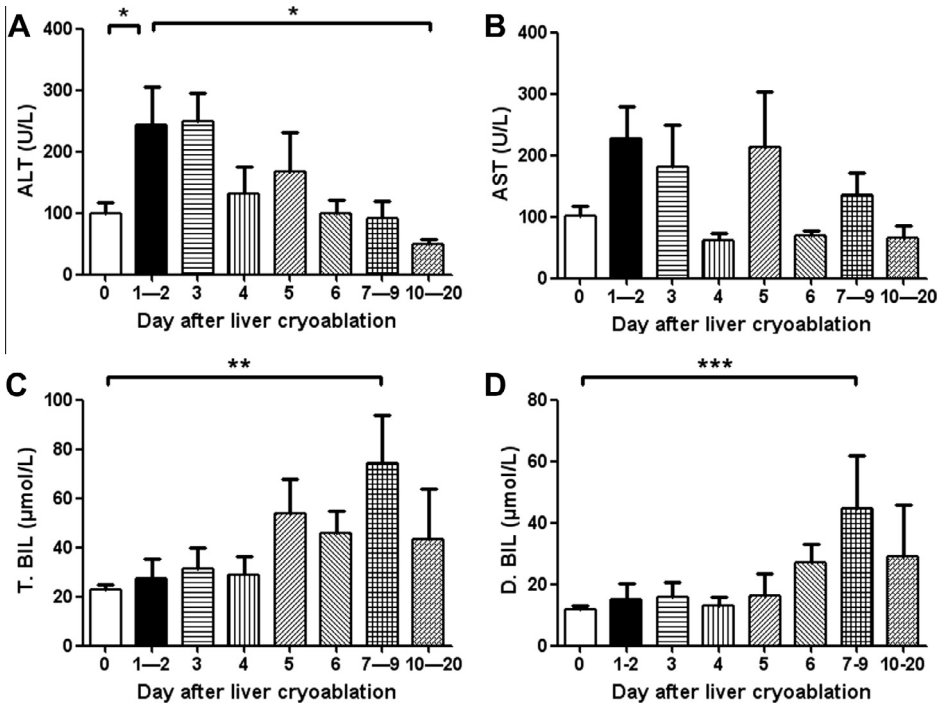

Fig. 3. Changes in the hepatic functional reserve of 14 hepatitis patients. Bonferroni's multiple comparison test was used to compare different days. The number of test results obtained varied between days (18, 8, 6, 4, 4, 5, 6 and 6 results were obtained on day 0, 1–2, 3, 4, 5, 6, 7–9 and 10–20, respectively); thus, days with fewer than three test results were merged with neighboring days. Indices of hepatic functional reserve were alanine transaminase (ALT) (A), aspartate transaminase (AST) (B), total bilirubin (T.BIL) (C) and direct bilirubin (D.BIL) (D). $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

sessions (5%) but resolved after blood transfusion. Among the 24 patients who had thrombocytopenia before liver cryoablation, the thrombocytopenia had disappeared after 19 sessions and coagulopathy occurred within 4 days after eight sessions; after administration of rhIL-11, all patients recovered within 1 week. Liver abscess was observed after six sessions (5%) at the site of cryoablation, but the patients recovered within 2 weeks with antibiotic and drainage treatment.

Changes of hepatic functional reserve after simple cryotherapy

Among the 38 subjects, 14 were hepatitis positive. Fifty-one cryosurgeries were performed in these patients; transaminase levels were abnormal before 39 of these ablations and bilirubin was abnormal before 18 ablations. The test results were as follows: ALT 61 ± 52 U/L; AST 81 ± 52 U/L; T.BIL 23 ± 13 μmol/L; and D.BIL 11 ± 7 μmol/L. In 3 days post cryosurgery, the serum transaminase and bilirubin values rose rapidly to a peak, then fell gradually; in the 26 hepatitis negative patients, 34 cryosurgeries were performed, the maximal elevating fold of ALT, AST, T.BIL and D.BIL were 13, 16, 4 and 3, respectively; in the 12 hepatitis positive patients, 17 cryosurgeries were performed, the maximal elevating fold of ALT, AST, T.BIL and D.BIL were 10, 12, 4 and 4, respectively.

Changes of hepatic functional reserve after cryoprotective therapy

Among the 51 subjects, 66 cryosurgeries were performed in these patients; transaminase levels were abnormal before 53 of these ablations and bilirubin was abnormal before 27 ablations. The test results were as follows: ALT 71 ± 41 U/L; AST 85 ± 37 U/L; T.BIL 24 ± 12 μmol/L; and D.BIL 13 ± 6 μmol/L. The day after cryosurgery, the serum transaminase values rose rapidly to a peak (ALT 241 ± 48 U/L; AST 427 ± 45 U/L; both $P < 0.001$) and then fell gradually; ALT and AST decreased significantly from day 5 and day 2, respectively (both $P < 0.001$) (Fig. 1A and B). No obvious change in serum bilirubin was observed until 20 days post-cryosurgery (Fig. 1C and D).

In the 37 hepatitis negative patients, 48 cryosurgeries were performed. Transaminase levels were abnormal before 35 of these ablations and bilirubin was abnormal before 19 ablations. The test results were as follows: ALT 61 ± 11 U/L; AST 79 ± 8 U/L; T.BIL 24 ± 2 μmol/L; and D.BIL 13 ± 2 μmol/L. Due to abnormal liver function, liver-protective therapy was given before 35 (73%) ablations. The day after cryosurgery, the serum transaminase values rose rapidly to a peak (ALT 239 ± 54 U/L; AST 437 ± 50 U/L; both $P < 0.001$) and then fell gradually; ALT and AST decreased significantly from day 5 ($P < 0.01$) and day 2 ($P < 0.001$) (Fig. 2A and B). No obvious change in serum bilirubin was observed until 20 days post-cryosurgery (Fig. 2C and D).

In the 14 hepatitis positive patients, 18 cryosurgeries were performed. No patient had normal transaminase levels and bilirubin was abnormal before eight of these 18 ablations. The test results were as follows: ALT 99 ± 19 U/L; AST 102 ± 15 U/L; T.BIL 23 ± 2 μmol/L; and D.BIL 12 ± 1 μmol/L. Due to abnormal liver function, all 14 patients were given liver-protective therapy before the ablations. Serum ALT rose to 243 ± 62 U/L in 2 days ($P < 0.05$) and then fell gradually; the level decreased significantly after day 10 (48 ± 9 U/L, $P < 0.05$; Fig. 3A). Serum AST fluctuated over time, with no significant difference between time points (Fig. 3B). Serum bilirubin rose gradually to a peak 7–9 days after cryosurgery (T.BIL: 74 ± 19 μmol/L, $P < 0.01$, Fig. 3C; D.BIL: 45 ± 17 μmol/L, $P < 0.001$, Fig. 3D) and then fell gradually.

Discussion

Cryosurgery is becoming an attractive technique for non-resectable hepatic tumors. The complications of cryoablation for HCC are acceptable and the therapy can prolong the patient's life, especially

in those who undergo multiple cryoablations, which can further improve survival [12]. For tumors of diameter >6 cm in this study, two or three sessions of cryosurgery were performed to avoid serious side effects. To minimize impairment of liver function, liver-protective therapy was given before and after the ablations. Based on clinical observations, 1 week's rest was usually sufficient for patients to recover their strength and for most tumor breakage products to be metabolized. All of the patients in this study tolerated the treatment and no severe complications occurred after cryoablation, demonstrating the safety and efficacy of this technique.

Cryosurgery for liver cancer inevitably induces hepatocyte destruction and the subsequent release of transaminases and bilirubin, which are important indices of hepatic functional reserve. From 1988 to 2005, much research was undertaken in the field of cryotherapy and all studies found obvious elevations of AST and ALT [6,7,17,20,26] and bilirubin (including T.BIL and D.BIL) [1,7,11]; these elevations are usually transient – especially in patients with a healthy liver – and usually normalize within a few days. In the present study, although most patients received liver-protective treatment before cryosurgery, transaminase levels were doubled (mean ALT and AST were 71 U/L and 85 U/L, respectively) compared with the normal ranges in our hospital. One day after cryosurgery, ALT and AST were increased 3.3-fold (peak mean was 241 U/L) and fivefold (peak mean was 427 U/L), respectively, but were close to the preoperative level 5 days post-procedure. Hamad and Neifeld [7] showed that ALT and AST were increased 13- and 15-fold, respectively, by postoperative day 1 and then gradually subsided. Compared with this study, our finding of increased AST and ALT is consistent, but the extent of the increase differs. The explanation for the lower increase in our study may be due to two factors: the time of measurement and the use of liver-protective therapy. Nair et al. [13] found that AST and ALT levels were increased after all procedures and peaked at 6 h, but we measured AST and ALT 24 h after cryosurgery. Other studies did not provide any liver protection before or after ablation, whereas in the present study, with the aid of liver-protective therapy, the rapid release of transaminases was controlled and recovered quickly. It seems that the use of liver-protective therapy is beneficial in cryoablation.

No significant increase of serum bilirubin was observed in the present study, which differs from the findings of Goodie et al. [6] and Hamad and Neifeld [7]. Goodie et al. demonstrated an increase in serum T.BIL from 11 μmol/L preoperatively to 28 μmol/L on postoperative day 1 and Hamad showed that serum T.BIL almost tripled on postoperative day 1. These differences may be explained by the fact that the patients in the present study were divided into hepatitis B virus positive and hepatitis B virus negative groups, and it was only in the hepatitis B positive group that T.BIL (74 μmol/L/23 μmol/L = 3.2) and D.BIL (45 μmol/L/12 μmol/L = 3.8) increased by more than 3 times 7–9 days after treatment. The reason for the serum bilirubin increase in hepatitis B positive patients remains unknown and needs further research.

Overall, the present results demonstrate that ALT and AST levels are important indicators of liver function following cryosurgery. In patients with hepatitis B virus, serum bilirubin increased by 3 times the preoperative level 7–9 days after cryosurgery. Liver-protective treatment may alleviate liver function damage following cryosurgery. In conclusion, we can say that cryosurgery for liver cancer is relatively safe in terms of liver function, but the research on long-term benefits are still needed in the future.

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