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ORIGINAL ARTICLE

Multimodal therapy for liver cirrhosis patients with advanced hepatocellular carcinoma

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Abstract
Purpose We have previously shown that continuous intra-arterial combination chemotherapy (IACC) might be more effective for advanced hepatocellular carcinoma (aHCC) in patients with HCV-related liver cirrhosis (C-LC) or alcoholic liver cirrhosis (A-LC) than in patients with HBV-related LC (B-LC). However, it is still unknown whether IACC actually improves the prognosis of aHCC patients with liver cirrhosis (LC), because it is difficult to perform a randomized controlled trial for patients with a poor prognosis. The aim of this study was to retrospectively assess the influence of IACC on the prognosis of aHCC.
Methods Fifty-eight adult Japanese patients who had aHCC and LC underwent repeated trans-arterial chemoembolization (TACE) without IACC between 1990 and 1997 at our hospital (group T), while 43 patients with aHCC and LC received IACC between 2000 and 2008 after undergoing several TACE sessions (group R). The Japan Integrated Staging score (JIS score) of each patient was ≥ 3 at the time of presentation, except for patients with tumor thrombi involving the first or subsequent portal vein branches or those with tumor invasion of the inferior vena cava. The same IACC regimen was repeated for as long as possible in group R.
Results In group T, 13 patients had B-LC, 37 patients had C-LC, and 8 patients had A-LC, while the respective numbers were 14, 21, and 8 in group R. The median survival time (MST) was 248 days for patients with C-LC in group T and 708 days for those in group R, while it was 253 days for patients with A-LC in group T and 593 days for those in group R. There were significant differences of survival between the two groups. However, MST was 369 days for patients with B-LC in group T and 782 days for those in group R, without a significant difference. In group R, a complete or partial response was achieved after 4 weeks of chemotherapy in 14.3% of patients with B-LC versus 42.9% of patients with C-LC and 37.5% of patients with A-LC.
Conclusions In LC patients with a JIS score > 3 at diagnosis, multimodal therapy with IACC after TACE prolongs the MST of C-LC or A-LC patients compared with TACE alone, although it does not improve the MST of patients with B-LC.

Keywords Multimodal therapy · Hepatocellular carcinoma · Liver cirrhosis · Intra-arterial chemotherapy · Trans-arterial chemoembolization

Introduction

Hepatocellular carcinoma (HCC) is the fifth most common malignancy in men and the eighth in women, with over 500,000 new cases being diagnosed worldwide each year [1–3]. Several therapeutic modalities, including surgery, percutaneous ethanol injection (PEI), transcatheter arterial chemoembolization (TACE), and radiofrequency ablation (RFA), are used to treat patients with small tumors. TACE has been reported to be an effective palliative treatment for patients with unresectable HCC, and various chemotherapy agents (such as doxorubicin, epirubicin, and mitomycin)

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are used with lipiodol [4–10]. Although repeated TACE is one of the most potent therapies for unresectable HCC, resistance often develops after a number of sessions, and long-term survival rates beyond 3 years are not very high.

There are also many patients with advanced HCC (aHCC), and intra-arterial chemotherapy is one of their few remaining options. It was reported that most patients with aHCC do not survive for longer than 6 months after diagnosis [11], while other authors have reported an average survival period of 4 months from the onset of symptoms and 2 months from the time of admission [12]. Development of better implantable drug delivery systems has made it possible to perform repeated hepatic arterial infusion of anticancer agents in patients with aHCC, and such hepatic arterial infusion therapy improves both survival and the quality of life [13]. Continuous local intra-arterial infusion of 5-fluorouracil (5-FU) and cisplatin (CDDP) via a pump and an implanted reservoir has been shown to prolong the survival of patients with aHCC [13–15]. We have also reported that the combination of low-dose intra-arterial 5-FU, CDDP, and leucovorin (LV) prolongs the survival of aHCC patients [16], with continuous infusion for 24 h being more effective than infusion for 6 h in aHCC patients with liver cirrhosis (LC) secondary to HCV infection, although 24-h infusion is associated with stronger hematologic toxicity [17]. Furthermore, we have demonstrated that IACC might be more effective for aHCC patients with alcoholic LC or HCV-related LC than for patients with HBV-related LC [18].

However, it is still unclear whether IACC is really useful for aHCC patients with LC, because the execution of a randomized controlled trial is very difficult in patients who have such a poor prognosis. Recently, sorafenib has also been used to treat patients with aHCC. Sorafenib is an oral multikinase inhibitor that shows strong in vitro activity by targeting the Raf/mitogen-activated protein kinase/extracellular signal-related kinase signaling pathway. Sorafenib inhibits cell surface tyrosine kinase receptors (vascular endothelial growth factor receptor [VEGFR]-2, VEGFR-3, and platelet-derived growth factor receptor-β) and downstream intracellular serine/threonine kinases (Raf-1, wild-type B-Raf, and mutant B-Raf) that are involved in both tumor cell proliferation and tumor angiogenesis [19]. Sorafenib has demonstrated significant clinical activity against HCC in phase II and phase III studies [20, 21], in which sorafenib treatment achieved a longer median survival time and longer time to radiologic progression compared with placebo. To devise future therapeutic strategies employing sorafenib for LC patients with aHCC, it is necessary to confirm the superiority of multimodal therapy with IACC after several sessions of TACE, compared to the effect of TACE alone. Accordingly, the aim of this study was to retrospectively assess the influence of IACC on the prognosis of patients with LC receiving TACE for aHCC.

Methods

Patients

Fifty-eight adult Japanese patients (53 men and 5 women) who had both aHCC and LC underwent repeated TACE without IACC between 1990 and 1997 at our hospital (group T). Forty-three patients (39 men and 4 women) who had aHCC and LC received IACC between 2000 and 2008 after several sessions of TACE (group R). In group R, patients were given IACC when TACE did not achieve a response or tumors showed rapid progression. The Japan Integrated Staging score (JIS score) of each patient was ≥ 3, and each patient had stage II or more advanced disease at the time of presentation (Table 1), except for patients with tumor thrombi involving the first or subsequent portal vein branches or those with tumor invasion of the inferior vena cava. The diagnosis of HCC was made either by histological examination or by a combination of imaging findings [double-contrast spiral computed tomography (CT)] and a serum alpha-fetoprotein (AFP) level > 500 μg/L. The patients in this study were not eligible for surgical resection or for interventions such as PEI or RFA because they had multiple tumors in both lobes of the liver. They also did not request liver transplantation.

Transcatheter arterial chemoembolization

All patients gave informed content to the procedure. A peripheral line was inserted for hydration. The femoral artery was catheterized under local anesthesia, and the catheter was advanced as close as possible to the tumor-feeding artery. Then, an emulsion of epirubicin and lipiodol followed by gelatin sponge particles was carefully

Table 1 Japan Integrated Staging score (JIS Score) calculation of JIS score

Variables	Score			
	0	1	2	3
Child-Pugh stage	A	B	C	
TNM stage*	I	II	III	IV

	Stage I	Stage II	Stage III	Stage IV
Child-Pugh C	2	3	4	5
Child-Pugh B	1	2	3	4
Child-Pugh A	0	1	2	3

* By Liver Cancer Group of Japan

injected under fluoroscopic monitoring. The emulsion contained 40 mg of epirubicin (Nipponkayaku Co., Tokyo, Japan) to each 5.0 ml of lipiodol. The doses of emulsion and gelatin particles were based on the size and number of the tumors.

TACE was performed in patients without main portal vein thrombus or severe liver dysfunction, while chemolipiodolalization was performed in patients with severe liver dysfunction.

IACC

Patients received 24-h IACC (LV at 12 mg/h, CDDP at 10 mg/h (Yakult Co., Tokyo, Japan) and 5-FU at 250 mg/m^2 for 22 h (Kyowa Kirin Co., Tokyo, Japan)). Continuous infusion was performed via the proper hepatic artery every 5 days for 4 weeks using a catheter connected to a subcutaneously implanted drug delivery system. After the first course, this regimen was repeated for as long as possible. The doses of the chemotherapy agents were selected according to previous reports [16, 17].

In all patients, an intra-arterial catheter was inserted via the femoral artery and was attached to a subcutaneous reservoir [22]. The gastroduodenal artery and the right gastric artery were occluded with steel coils to prevent gastroduodenal injury by the anticancer agents. Written informed consent to IACC was obtained from all of the patients.

Evaluation of response

The primary endpoint of this study was the response rate, which was evaluated according to World Health Organization (WHO) criteria. The secondary endpoints were progression-free survival and the toxicity of chemotherapy.

On CT scans obtained after 4 weeks of treatment, the size of the liver tumors was measured as the product of the two longest perpendicular diameters of the largest tumor. A complete response (CR) was defined as disappearance of the tumor, while a partial response (PR) was defined as more than 50% reduction in the product of the two longest diameters. An increase in the product by more than 25% was defined as progressive disease (PD), and changes between PD and PR were defined as stable disease (SD). Survival was measured as the interval between the date starting of IACC and death.

Statistical analysis

Statistical analysis was performed with the Statistical Package for the Social Science (SPSS version 11.0; SPSS, Chicago, IL, USA). Survival was evaluated by the Kaplan–Meier method, and the significance of differences in survival was determined by the log-rank test. Proportional hazards analysis was employed to identify factors with an influence on the curative effect and survival. Results are reported as the mean ± SD. A probability value of less than 0.05 was considered to indicate statistical significance.

Results

In group T, there were 13 patients with underlying HBV infection (B-LC), 37 patients with underlying HCV infection (C-LC), and 8 patients with alcoholic cirrhosis (A-LC), while the respective numbers were 14, 21, and 8 in group R. The Child-Pugh (C-P) class was A for 15 out of 58 patients in group T, while 36 were in class B and 7 were in class C, with the respective numbers being 19, 20, and 4 in group R (Table 2). The median survival time was 238 days for group T and 680 days for group R (Fig. 1). But there was no significant difference of the median survival between the two groups ($P = 0.956$, by the Kaplan–Meier method and log-rank test).

Table 2 Clinical characteristics of 101 patients with advanced HCC and cirrhosis

	Group T	Group R	*P*
No. of patients	58	43	
Mean age	62.3 ± 7	62.8 ± 7	0.437
Gender (M/F)	53/5	39/4	0.906
Type of cirrhosis (HBV/HCV/alcohol)	13/37/8	14/21/8	0.629
Child-Pugh classification (A/B/C)	15/36/7	19/20/4	0.052
Stage (II/III/IVA/IVB)	4/9/41/4	0/5/32/6	0.076
JIS score (3/4/5)	25/25/8	26/13/4	0.097

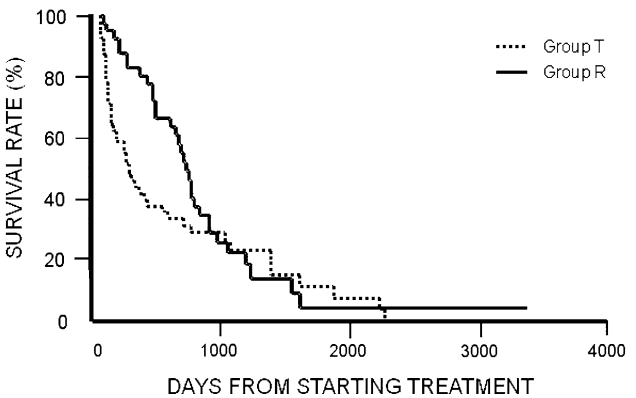

Fig. 1 Survival curves for LC patients with aHCC plotted by the Kaplan–Meier method. There were 58 patients treated with TACE alone (group T) and 43 patients treated with IACC after TACE (group R). The survival of group R was better than that of group T, although there was no significant difference. The median survival time was 238 and 680 days for group T and group R, respectively ($P = 0.956$, Kaplan–Meier method and log-rank test)

Table 3 Objective responses of 43 patients with HOC and cirrhosis treated by combined intra-arterial chemotherapy

	CR	PR	SD	PD	Response rate (%)	*P*
HBV $n = 14$	0	2	3	9	14.3	
HCV $n = 21$	0	9	7	5	42.9	0.755
Alcohol $n = 8$	0	3	5	0	37.5	0.873

Data show the number of patients

CR complete response, *PR* partial response, *SD* stable disease, *PD* progressive disease

Table 4 Clinical characteristics of the 27 patients with advanced HCC- and HBV-related cirrhosis

	Group T	Group R	*P*
No. of patients	13	14	
Mean age	55.5 ± 6	59.9 ± 10	0.105
Gender (M/F)	12/1	14/0	0.756
Child-Pugh classification (A/B/C)	4/7/2	8/5/1	0.220
Stage (II/III/IVA/IVB)	0/3/9/1	0/1/10/3	0.280
JIS score (3/4/5)	7/4/2	10/31	0.430

We previously demonstrated that IACC might be more effective for aHCC in patients with A-LC or C-LC than in patients with B-LC [18]. In this retrospective study, the JIS score of each patient was already 3 or more at presentation, except for patients with tumor thrombi involving the first or subsequent portal vein branches or those with tumor invasion of the inferior vena cava. A complete or partial response was seen after 4 weeks of chemotherapy in only 14.3% of patients with B-LC versus 42.9% of patients with C-LC and 37.5% of patients with A-LC from group R (Table 3). Therefore, we investigated the usefulness of IACC in relation to the etiology of aHCC.

Assessment stratified by the etiology of cirrhosis

HBV-related cirrhosis

The C-P class was A for 4 out of 13 patients with HBV-related cirrhosis from group T, while 7 were in class B and 2 were in class C, with the respective numbers being 8, 5, and 1 in group R (Table 4). There was no significant difference of survival between the groups ($P = 0.615$, Kaplan–Meier method and log-rank test), although the median survival time was only 369 days for the patients in group T versus 782 days for those in group R (Fig. 2). The 1-year survival rate of the patients in group T was 46.2%, while it was 57.1% for those in group R. The 2-year survival rate of patients in group T was 23.1 versus 50.0% for those in group R, while 5-year survival rate was 7.7 and 7.1%, respectively (Table 5).

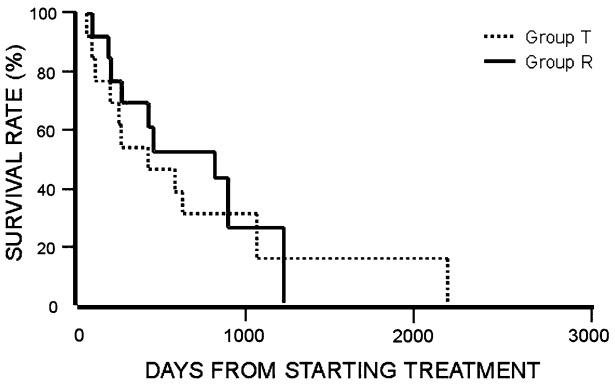

Fig. 2 Survival curves for B-LC patients with aHCC plotted by the Kaplan–Meier method. There were 13 patients treated with TACE alone (group T) and 14 patients treated with IACC after TACE (group R). There was no significant difference of survival between groups T and R, with the median survival time being 369 and 782 days, respectively ($P = 0.615$, Kaplan–Meier method and log-rank test)

Table 5 Survival in relation to the etiology in patients with HCC and cirrhosis

Survival rate (%)	Etiology		
	HBV	HCV	Alcohol
1 year			
TACE	46.2	35.1	25.0
TACE + IACC	57.1	90.5	50.0
2 years			
TACE	23.1	24.3	0.0
TACE + IACC	50.0	47.6	25.0
3 year			
TACE	15.4	16.2	0.0
TACE + IACC	21.4	19.0	0.0
5 years			
TACE	7.7	5.4	0.0
TACE + IACC	7.1	9.5	0.0

HCV-related cirrhosis

The C-P class was A for 9 out of 37 patients with HCV-related cirrhosis from group T, while 24 were in class B and 4 were in class C, with the respective numbers being 10, 10, and 1 in group R (Table 6). The median survival time was 248 days for the patients in group T versus 708 days for those in group R, and there was significant difference between the groups ($P = 0.045$, Kaplan–Meier method and log-rank test) (relative hazard: 0.56; 95% confidence interval: 0.31–0.99 [$P = 0.048$]) (Fig. 3). The 1-year survival rate of the patients in group T was 35.1%, while it was 90.5% for those in group R. The 2-year survival rate of patients in group T was 24.3 versus 47.6% for those in group R, while the 5-year survival rate was 5.4 and 9.5%, respectively (Table 5).

Table 6 Clinical characteristics of 58 patients with advanced HCC- and HCV-related cirrhosis

	Group T	Group R	*P*
No. of patients	37	21	
Mean age	65.2 ± 4	65.9 ± 5	0.126
Gender (M/F)	33/4	17/4	0.386
Child-Pugh classification (A/B/C)	9/24/4	10/10/1	0.068
Stage (ll/lll/IVA/IVB)	3/5/27/2	0/4/15/2	0.564
JIS score (3/4/5)	15/19/3	14/6/1	0.067

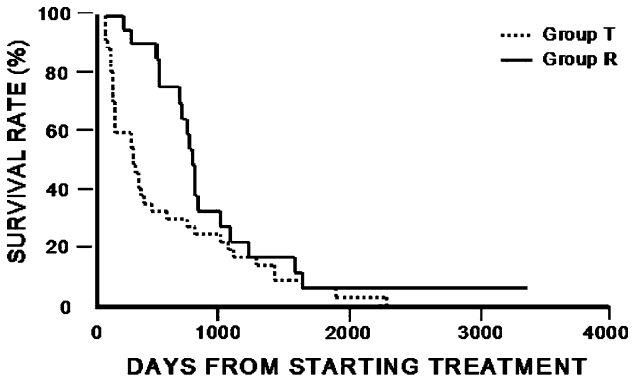

Fig. 3 Survival curves for C-LC patients with aHCC plotted by the Kaplan–Meier method. There were 37 patients treated with TACE alone (group T) and 21 patients treated with IACC after TACE (group R). The survival of group R was significantly better than that of group T, with the median survival time being 248 and 708 days, respectively. ($P = 0.045$, Kaplan–Meier method and log-rank test) (relative hazard: 0.56; 95% confidence interval: 0.31–0.99 [$P = 0.048$])

Table 7 Clinical characteristics of 16 patients with advanced HCC and alcoholic cirrhosis

	Group T	Group R	*P*
No. of patients	8	8	
Mean age	59.9 ± 8	61.1 ± 3	0.328
Gender (M/F)	8/0	8/0	1.000
Child-Pugh classification (A/B/C)	2/4/2	1/5/2	0.798
Stage (ll/lll/IVA/IVB)	1/1/5/1	0/0/7/1	0.505
JIS score (3/4/5)	3/2/3	1/5/2	0.798

Alcoholic cirrhosis

The C-P class was A for 2 out of 8 patients with alcohol cirrhosis from group T, while 4 were in class B and 2 were in class C, with the respective numbers being 1, 5, and 2 in group R (Table 7). The median survival time was 253 days for the patients in group T and 593 days for those in group R, while a significant difference between the groups ($P = 0.025$, Kaplan–Meier method and log-rank test) (relative hazard: 0.19; 95% confidence interval: 0.04–0.95 [$P = 0.044$]) (Fig. 4). The 1-year survival rate of the patients in group T was 25.0%, while it was 50.0% for those in group R. The 2-year survival rates were 0.0 and 25.0%, respectively (Table 5).

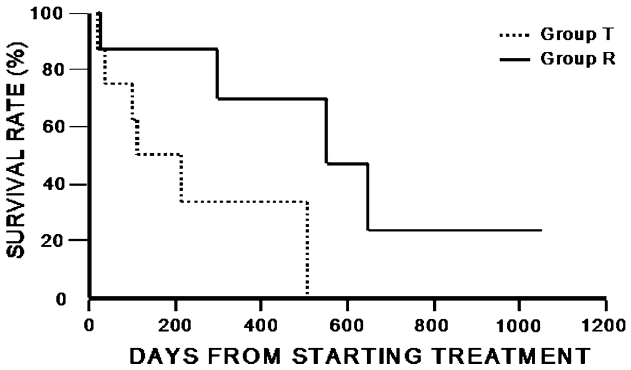

Fig. 4 Survival curves for A-LC patients with aHCC plotted by the Kaplan–Meier method. There were 8 patients treated with TACE alone (group T) and 8 patients treated with IACC after TACE (group R). The survival of group R was significantly better than that of the group T, with the median survival time of each group being 253 and 593 days, respectively ($P = 0.025$, Kaplan–Meier method and log-rank test) (relative hazard: 0.19; 95% confidence interval: 0.04–0.95 [$P = 0.044$])

Discussion

TACE has been widely used for the palliative treatment of HCC that is unsuitable for curative therapy. Randomized controlled trials and prospective studies have shown that TACE improves the survival of patients with unresectable HCC [8–10]. However, a single session of TACE does not always achieve complete necrosis of the tumor [23], and repetition of this treatment has been recommended [7]. Frequent recurrence of aHCC after TACE is caused by tumor angiogenesis and incomplete embolism. IACC improves both the survival of patients with aHCC and their quality of life [13]. We have already reported that IACC prolongs the survival of patients with aHCC [16] and that it might be more effective for patients with A-LC or C-LC than for those with B-LC [18]. To devise future therapeutic strategies including sorafenib, it is necessary to confirm whether multimodal therapy (IACC after TACE) is more useful for LC patients with aHCC than TACE alone.

In the present study, we retrospectively assessed the effect on survival of IACC after TACE for patients with aHCC stratified by the etiology of LC. There was no significant difference of survival between group T and group R, although the median survival time was 238 days for group T and 680 days for group R. However, stratified analysis based on the etiology of LC showed that adding IACC to TACE prolonged the median survival of C-LC or A-LC patients compared with TACE alone, although it did not improve the median survival of patients with B-LC.

When the percentage of patients with a complete or partial response after 4 weeks of chemotherapy was assessed, IACC was also more effective for aHCC in patients with C-LC or A-LC than in patients with B-LC. These results indicate that adding IACC after TACE can improve the survival of C-LC and A-LC patients (but not B-LC patients) with aHCC compared to TACE alone and that prolongation of survival by multimodal therapy depends on the response to IACC. Indeed, C-LC and A-LC patients showed a good response to IACC compared with B-LC patients and had superior 1-year and 2-year survival rates. However, patients with A-LC treated by multimodal therapy did not achieve long survival, probably because their cirrhosis progressed due to continued alcohol intake.

Why there were differences in the response of aHCC to IACC depending on the etiology of LC, although the same chemotherapy regimen was used? When chemotherapy is given to LC patients with aHCC, we must consider the influence of tumor factors, the host immune system, and anticancer drugs. Production of immunosuppressive factors, an increase in regulatory T cells (Treg), and down-regulation of tumor antigens and MHC molecules are mechanisms by which tumor cells escape the host immune response [24, 25]. These mechanisms all operate in patients with HCC. It has been demonstrated that HCC progresses despite the detection of a tumor-specific cellular and humoral immune response in more than 50% of patients [26]. The response of T cells to self- and nonself-antigens is controlled by a network of regulatory T (Treg) cells. $CD4^+$ cells with constitutive expression of CD25, the interleukin-2-receptor α-chain, are generally considered to be natural Treg cells and account for 5–10% of peripheral $CD4^+$ T cells in healthy animals and humans [27–29]. Based on their cytokine profiles, helper T cells can be divided into two distinct populations, which are called type 1 helper T (Th1) cells and type 2 helper T (Th2) cells. Th1 cells produce interferon-γ (IFN-γ) and interleukin-2 (IL-2) and play a pivotal role in cell-mediated immunity, while Th2 cells produce interleukin-4 (IL-4), interleukin-10 (IL-10), and other cytokines that are essential for the regulation of humoral immunity [30, 31]. IFN-γ preferentially inhibits the proliferation of Th2 cells, while IL-4 and IL-10 are secreted by Th2 cells and inhibit the secretion of IL-12, which is the critical cytokine for Th1 differentiation [32, 33]. In addition, Th1 and Th2 cells cross-regulate their own development. It has been reported that Th2 cytokines down-regulate antitumor immunity [34], while the activation of the Th1 response promotes antitumor immunity [35–38]. We previously reported that the Th1/Th2 balance might be a useful indicator of the response of aHCC to IACC in patients with LC and that loss of Th1 dominance is significantly more common in patients with tumor progression [39]. In addition, the percentage of Th2 cells was reported to increase in LC patients with aHCC as the response to IACC decreases [40]. Thus, the Th1/Th2 balance might be titled toward Th2 predominance in patients with B-LC, and this may help to explain why IACC was more effective for aHCC in patients with C-LC or A-LC than in those with B-LC.

In conclusion, we studied LC patients with a HCC and a JIS score ≥ 3 at presentation. We found that multimodal therapy with IACC after TACE prolonged the survival compared with TACE alone in patients with C-LC or A-LC, although it did not improve the survival for patients with B-LC. The improvement of survival by adding IACC to TACE in C-LC or A-LC patients with aHCC was dependent on the response to IACC. Further studies are needed to improve the survival of B-LC patients with aHCC.

References

1. Parkin DM, Bray F, Ferlay J et al (2001) Estimating the world cancer burden: Globocan 2000. Int J Cancer 94:153–156
2. Yu MC, Yuan J-M (2004) Environmental factors and risk for hepatocellular carcinoma. Gastroenterology 127:S72–S78
3. Bosch FX, Ribes J, Diaz M et al (2004) Primary liver cancer: worldwide incidence and trends. Gastroenterology 127:S5–S16
4. Chuang VP, Wallace S (1981) Hepatic arterial embolization in the treatment of hepatic neoplasms. Radiology 140:51–58
5. Yamada R, Sato M, Kawabata M et al (1983) Hepatic artery embolization in 120 patients with unresectable hepatoma. Radiology 148:397–401
6. Lin DY, Liaw YF, Lee TY et al (1988) Hepatic arterial embolization in patients with unresectable hepatocellular carcinoma: a randomized controlled trial. Gastroenterology 94:453–456
7. Ikeda K, Kumada H, Saitoh S et al (1991) Effect of repeated transcatheter arterial embolization on the survival time in patients with hepatocellular carcinoma. Cancer 68:2150–2154
8. Llovet JM, Real MI, Montana X et al (2002) Barcelona liver cancer group. Arterial embolization or chemoembolization versus systemic treatment in patients with unresectable hepatocellular carcinoma: a randomized controlled trial. Lancet 359:1734–1739
9. Lo CM, Ngan H, Tso WK et al (2002) Randomized controlled trial of transarterial lipiodol chemoembolization for unresectable hepatocellular carcinoma. Hepatology 35:1164–1171
10. Takayasu K, Arii S, Ikai I et al (2006) Liver cancer study group of Japan. Prospective cohort study of transarterial chemoembolization for unresectable hepatocellular carcinoma in 8,510 patients. Gastroenterology 131:461–469
11. Okuda K, Ohtsuki T, Obata H, Tomimatsu M, Okazaki N, Hasegawa H, Nakajima Y, Onishi K (1985) Natural history of hepatocellular carcinoma and prognosis in relation to treatment. Study of 850 patients. Cancer 56:918–928
12. Nagasue N, Yukaya H, Hamada T, Hirosue S, Kanashima R, Inokuchi K (1984) The natural history of hepatocellular carcinoma. A study of 100 untreated cases. Cancer 54:1461–1465
13. Toyoda H, Nakano S, Kumada T, Takeda I, Sugiyama K, Osada T, Kirishima S, Suga T, Takahashi M (1995) The efficacy of continuous local arterial infusion of 5-fluorouracil and cisplatin through an implanted reservoir for severe advanced hepatocellular carcinoma. Oncology 52:295–299

14. Murata K, Shiraki K, Kawakita T, Yamamoto N, Okano H, Nakamura M, Takahisa Sakai, Deguchi M, Ohmori S, Nakano T (2003) Low-dose chemotherapy of cisplatin and 5-fluorouracil or doxorubicin via implanted fusion port for unresectable hepatocellular carcinoma. Anticancer Res 23:1719–1722
15. Okuda K, Tanaka M, Shibata J, Ando E, Ogata T, Kinoshita H, Eriguchi N, Aoyagi S, Takikawa K (1999) Hepatic arterial infusion chemotherapy with continuous low dose administration of cisplatin and 5-fluorouracil for multiple recurrence of hepatocellular carcinoma after surgical treatment. Oncology Report 6:587–591
16. Nagai H, Sumino Y (2008) Therapeutic strategy of advanced hepatocellular carcinoma by using combined intra-arterial chemotherapy. Recent Pat Anticancer Drug Discov 3:220–226
17. Nagai H, Kanayama M, Higami K et al (2007) Twenty-four hour intra-arterial infusion of 5-fluorouracil, cisplatin, and leucovorin is more effective than 6-hour infusion for advanced hepatocellular carcinoma. World J Gastroenterol 13(2):280–284
18. Kanayama M, Nagai H, Sumino Y (2009) Influence of the etiology of liver cirrhosis on the response to combined intra-arterial chemotherapy in patients with advanced hepatocellular carcinoma. Cancer Chemother Pharmacol 64:109–114
19. Wilhelm SM, Carter C, Tang L et al (2004) BAY 43–9006 exhibits broad spectrum oral antitumor activity and targets the RAF/MEK/ERK pathway and receptor tyrosine kinases involved in tumor progression and angiogenesis. Cancer Res 64:7099–7109
20. Moreno-Aspitia A, Morton RF, Hillman DW et al (2009) Phase II trial of sorafenib in patients with metastatic breast cancer previously exposed to anthracyclines or taxanes: North Central cancer treatment group and Mayo Clinic Trial N0336. J Clin Oncol 27:11–15
21. Abou-Alfa GK, Schwartz L, Ricci S et al (2006) Phase II study of sorafenib in patients with advanced hepatocellular carcinoma. J Clin Oncol 24:4293–4300
22. Iwamiya T, Sawada S, Ohta Y (1994) Repeated arterial infusion chemotherapy for inoperable hepatocellular carcinoma using an implantable drug delivery system. Cancer Chemother Pharmacol 33:S134–S138
23. Wakasa K, Sakurai M, Kuroda C et al (1990) Effect of transcatheter arterial embolization on the boundary architecture of hepatocellular carcinoma. Cancer 65:913–919
24. Gabrilovich DI, Chen HL, Girgis KR et al (1996) Production of vascular endotherial growth factor by human tumors inhibits the functional maturation of dendritic cells. Nat Med 2:1096–1103
25. Ferrara N, Gerber HP, LeCounter J (2003) The biology of VEGF and its receptors. Nat Med 9:669–676
26. Korangy F, Ormandy LA, Bleck JS et al (2004) Spontaneous tumor-specific humoral and cellular immune response to NY-ESO-1 in hepatocellular carcinoma. Clin Cancer Res 10:4332–4341
27. Shevach EM (2001) Certified professionals: $CD4^+$ $CD25^+$ suppressor T cell. J Exp Med 193:F41–F46
28. Stephens LA, Mottet C, Mason D, Powrie F (2001) Human CD4 + CD25 + thymocytes and peripheral T cells have immune suppressive activity in vitro. Eur J Immunol 31:1247–1254
29. Jonuleit H, Schmitt E, Stassen M, Tuettenberg A, Knop J, Enk AH (2001) Identification and functional characterization of human CD4 + CD25 + T cells with regulatory properties isolated from peripheral blood. J Exp Med 193:1285–1294
30. Mosmann TR, Coffman RL (1989) Th1 and Th2 cells: different patterns of lymphokine secretion lead to different functional properties. Annu Rev Immunol 7:145–173
31. O'Garra A (1998) Cytokines induce the development of functionally heterogeneous T helper cell subsets. Immunity 8:275–283
32. Roitt I (ed) (1997) Essential immunology, 9th edn. Blackwell Science, Malden
33. Kuby J, Cameron J, Todd C, Mitchell J (2000) Immunology, 4th edn. Freeman and Company, New York
34. Kobayashi M, Kobayashi H, Polland RB et al (1998) A pathogenic role of Th2 cells and their cytokine products on the pulmonary metastasis of murine B16 melanoma. J Immunol 160:5869–5873
35. Zitvogel L, Mayordomo JI, Tjandrawan T et al (1996) Therapy of murine tumors with tumor peptide-pulsed dendritic cells: dependence on T cells, B7 costimulation, and T helper cell 1-associated cytokines. J Exp Med 183:87–97
36. Tsung K, Meko JB, Peplinski GR et al (1997) IL-12 induces T helper 1-directed antitumor response. J Immunol 158:3359–3365
37. Weiner GJ, Liu HM, Wooldridge JE et al (1997) Immunostimulatory oligodeoxynucleotides containing the CpG motif are effective as immune adjuvants in tumor antigen immunization. Proc Natl Acad Sci USA 94:10833–10837
38. Hu H-M, Urba WJ, Fox BA (1998) Gene-modified tumor vaccine with therapeutic potential shifts tumor-specific T cell response from Type 2 to a type 1 cytokine profile. J Immunol 161:3033
39. Nagai H, Miyaki D, Matsui T, Kanayama M, Higami K, Momiyama K, Ikehara T, Watanabe M, Sumino Y, Miki K (2008) Th1/Th2 balance: an important indicator of efficacy for intra-arterial chemotherapy. Cancer Chemother Pharmacol 62:959–963
40. Momiyama K, Nagai H, Sumino Y (2009) Changes of host immunity in relation to efficacy in liver cirrhosis patients with advanced hepatocellular carcinoma treated by intra-arterial chemotherapy. Cancer Chemother Pharmacol 64:271–277