Most of the HCC patients had hepatitis or cirrhosis with underlying liver disorder and a reduction in hepatic blood flow to various degrees. Liver dysfunction in patients with HCC may affect the PK of sorafenib. When comparing the PK by Child–Pugh classification, geometric means of $AUC_{0\text{-}12}$ and C_{max} at steady state were lower in the Child–Pugh B group than in the Child–Pugh A group, whereas after multiple doses of sorafenib, the mean plasma concentrations were highly variable and showed no clear dose dependency. Although the numerical differences in geometric means for PK parameters such as AUC, C_{max}, and $t_{1/2}$ were observed between Child–Pugh classifications, these differences were considered not to be clinically relevant in consideration of their large intersubject variability. No significant difference in clinical findings between these two groups was observed. There was also no major difference (i.e. over 20%) in the incidence of adverse events between Child–Pugh A and B groups. However, geometric means of $AUC_{0\text{-}12}$ and C_{max} at steady state were slightly lower in the Child–Pugh B patients compared with the Child–Pugh A patients.

There were no remarkable differences in the overall incidence of adverse events for each dose level (92% for the 200-mg group and 100% for the 400-mg group). For a few drug-related adverse events, the incidences were at least 20% higher in the 400-mg group than in the 200-mg group, including rash or desquamation (71.4 *vs* 38.5%), hand–foot skin reaction (57.1 *vs* 30.8%), pruritus (50.0 *vs* 7.7%), decrease of platelets (35.7 *vs* 7.7%), hypertension (28.6 *vs* 7.7%), dry skin (21.4 *vs* 0%), and stomatitis or pharyngitis (21.4 *vs* 0%). DLT of hand–foot skin reaction was observed in a patient with Child–Pugh B at the end of cycle 1 with 400 mg bid, whereas no DLT was observed in the 200-mg bid group.

The most common drug-related adverse events were elevated lipase (88.9%) and amylase (59.3%). Twenty-four (88.9%) of the 27 patients showed high values of grade 3 or worse. Most of the patients were asymptomatic and only one patient had abdominal pain with findings to indicate pancreatitis on ultrasonography during cycle 6. His pancreatitis resolved shortly after discontinuation of sorafenib, and the patient restarted and continued with a reduced dose of sorafenib after recovery.

A separate phase I clinical study was carried out to evaluate the safety of sorafenib in patients with solid tumor, excluding HCC, at doses of 100, 200, 400, and 600 mg bid.[18] In that study, the most common type of adverse events included skin reaction, elevation of pancreatic enzyme, and gastrointestinal (GI) toxicity such as diarrhea. In the current study, a similar pattern of adverse events was observed. These results suggest that 'gastrointestinal' and 'dermatology/skin' are common adverse events regardless of cancer type and liver function status. One finding to note is that the incidence of elevation (grade 3/4) of lipase (63.0%) or amylase (14.8%) in the present study in HCC patients was higher than that observed in non-HCC patients (lipase 23% and amylase 10%).[18]

In summary, the present study showed no clinically significant difference in PK, safety, tolerability, or efficacy by Child–Pugh status or between HCC patients and non-HCC patients, whereas some dose dependency in adverse events was observed.

Investigations into cytotoxic agents for HCC have been conducted.[20,21] However, no standard chemotherapy has been established. Recently, a number of agents targeting growth factors were investigated in HCC. Through these investigations, it was indicated that epidermal growth factor receptor/human epidermal growth factor receptor 1 (EGFR/HER1) is actively expressed in human hepatoma cells.[22,23] Erlotinib, which is an EGFR/HER1 tyrosine kinase inhibitor, and lapatinib, which is an EGFR/HER1 and ErbB-2 (Her2/neu) dual tyrosine kinase inhibitor, have been investigated in phase II studies in HCC patients.[24–26] For erlotinib, the response rate was 4–9%, the median TTP was 2.1–3.2 months, and the OS was 5.8–13 months,[24,25] whereas for lapatinib, the response rate was 0%, and the median progression-free survival time was 1.8 months.[26]

Hepatocellular carcinoma, given its hypervascular characteristics, may be sensitive to antiangiogenic agents.[9] It is known that VEGF augments the development and metastasis of HCC. Bevacizumab, a monoclonal antibody against VEGF, has been investigated in phase II studies.[27] The response rate with bevacizumab was 10% and the disease control rate was 80%. A combination of gemox (gemcitabine plus oxaliplatin) and bevacizumab showed a better response rate of 20%.[28]

Sorafenib, an orally active multikinase inhibitor, blocks tumor-cell proliferation by targeting Raf/MEK/ERK signaling at the level of Raf kinase, and exerts an antiangiogenic effect by targeting VEGFR-2, VEGFR-3, and PDGFR-β tyrosine kinases. In phase II studies in non-Japanese and Japanese HCC patients, comparable median TTP of 4.2 and 4.9 months, respectively, and response rates of 2 and 4%, respectively, were shown.[15] However, OS in the two studies were different: 9.2 months in the non-Japanese study and 15.6 months in the Japanese study. Difference in backgrounds such as liver function or treatment after progression may play a role in this discrepancy in survival time.

In the current study, one patient achieved partial response (Fig. 1). The patient had several small viable HCC lesions after hepatectomy, percutaneous ethanol injection, and TACE. Following administration of sorafenib, tumor vascularity decreased dramatically preceding a gradual tumor reduction. Time to tumor shrinking varied across lesions, ranging from 1 to 8 months after initiation of treatment with sorafenib. It is likely that, with anti-VEGF agents such as sorafenib, it may take time to achieve tumor reduction to meet partial response by RECIST, whereas the duration of stable disease may persist due to its tumor stabilization activity.

With the relatively long TTP of VEGF pathway-targeting agents such as bevacizumab or sorafenib, these agents may have antitumor effects on HCC and prolong survival. With its profile of tumor stabilization and tolerability, sorafenib may be applicable not only for advanced HCC but also for the adjuvant setting after curative treatment, such as surgery or radiofrequency ablation therapy.

In conclusion, in the present phase I study, sorafenib demonstrated favorable safety and tolerability, and promising preliminary antitumor activity in Japanese HCC patients. Considering that DLT was observed in one of 14 patients treated with 400 mg bid, 400 mg bid could also be recommended for future studies in Japanese HCC patients, as well as non-HCC Japanese and Caucasian patients. However, as the number of patients was limited in this phase I study, a confirmatory study will be required with a larger number of patients.

Acknowledgments

This study was supported by Bayer Yakuhin.

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Annals of Surgical Oncology **15**(4):1008–1014
DOI: 10.1245/s10434-007-9705-0

Systemic Therapy for Hepatocellular Carcinoma: Cytotoxic Chemotherapy, Targeted Therapy and Immunotherapy

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Conventional cytotoxic chemotherapy has not provided clinical benefit or prolonged survival for patients with advanced HCC. This review summarizes the results of prospective clinical trials of several categories of systemic therapy, with emphasis on the more promising results from recent trials of biologically targeted therapeutic agents in HCC.

Key Words: Hepatocellular—Hepatoma—Chemotherapy—Chemoresistance—Clinical trials—Biologic therapy.

Hepatocellular carcinoma (HCC) is currently the fifth most common solid tumor worldwide, and the fourth leading cause of cancer-related death.[1] It is a lethal disease, as the annual incidence roughly equals the annual mortality.[2,3] Eighty percent of new cases occur in developing countries, but the incidence is rising in economically developed regions including Japan, Western Europe, and the United States.[4] More than 80% of patients present with advanced or unresectable disease, and for those patients who do undergo resection, the recurrence rates can be as high as 50% at 2 years.[5–7] Thus, many patients will seek systemic therapy. A 1997 meta-analysis evaluating the results of 37 randomized clinical trials of systemic and regional chemotherapy in 2803 HCC patients concluded that nonsurgical therapies were ineffective or minimally effective at best.[8] Most HCC patients have underlying cirrhosis and hepatic dysfunction, which complicates safely administering systemic therapy and conducting trials of new agents in this patient population.

Received June 1, 2007; accepted October 23, 2007; published online January 31, 2008.
Proceedings of the 5th International Meeting Hepatocellular Carcinoma: Eastern and Western Experiences held in Houston, TX, January 11–13, 2007.
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CYTOTOXIC CHEMOTHERAPY FOR HCC: REASONS FOR LACK OF EFFICACY

Most published studies of systemic chemotherapy report response rates of 0% to 25%, and there is no published evidence that systemic chemotherapy improves overall survival in any subset of HCC patients.[9–11] HCC comprises clinically chemotherapy-resistant tumors, and this observation is supported by low response rates across a wide variety of cytotoxic agents (Table 1). The most widely used agent has been doxorubicin, both as a single agent and in

TABLE 1. *Selected clinical trials in patients with advanced hepatocellular carcinoma*

Study	Regimen	Phase	Sample size	Response rate (%)	Median survival (mo)
Cytotoxic chemotherapy					
Yeo et al.[15]	PIAF vs. Adriamycin	3	94/94	20.9 vs. 10.5	8.6 vs. 6.83
Mok et al.[66]	Nolatrexed vs. doxorubicin	2	37/17	0	4.9 vs. 3.7
Posey et al.[67]	TI38067 vs. Adriamycin	2/3	169/170	NA	5.7 vs. 5.6
Gish et al.[16]	Nolatrexed vs. doxorubicin	3	444	1.4 vs. 4.0	5.5 vs. 8 (P = .0068)
Patt et al.[68]	Thalidomide	2	37	6%	6.8
Pastorelli et al.[69]	Pegylated doxorubicin + gemcitabine	2	35	23%	8.8
Immunotherapy/hormonal therapy					
Barbare et al.[70]	Tamoxifen vs. BSC	2	210/210	NA	4.8 vs. 4.0
O'Neil et al.[71]	Octreotide LAR	2	17		TTTF 3.5; OS 10
Lee et al.[61]	Dendritic cells	2	31	12.9%	1-y survival 40%
Targeted biologic therapy					
Llovet et al.[33]	Sorafenib vs. placebo	3	602	2.3%	10.7 vs. 7.9 (P = .00058)
Abou-Alfa et al.[32]	Sorafenib	2	137	2.2	9.3
Philip et al.[44]	Erlotonib	2	38	9%	13
Thomas et al.[43]	Erlotonib	2	40	0%	10.75
Thomas et al.[50]	Bevacizumab + erlotonib	2	34	20.6%	19 (PFS 9)
Zhu et al.[72]	Cetuximab	2	30	0%	PFS 6 wk; OS 22 wk
O'Dwyer et al.[73]	Gefitinib	2	31	3%	PFS 2.8; OS 6.5
Combination cytotoxic + biologic therapy					
Sun et al.[74]	Capecitabine, oxaliplatin, bevacizumab	2	30	11%	PFS 5.4
Zhu et al.[75]	Gemox + bevacizumab	2	33	20	9.6

PIAF, cisplatinum, interferon, doxorubicin, and 5-fluorouracil; OS, overall survival; PFS, progression-free survival; TTTF, time to treatment failure.

combination with other drugs.[12–14] An early randomized trial against best supportive therapy showed greatly increased survival, but this was only in the order of weeks. A pivotal phase 3 trial of doxorubicin combination chemotherapy (cisplatinum, interferon, doxorubicin, and 5-fluorouracil, PIAF) showed a statistically significant difference in response rate favoring PIAF, but no survival difference.[15] Another study in which doxorubicin was the control arm in a randomized phase 3 trial against nolatrexed showed a highly statistically significant survival benefit in favor of the control doxorubicin arm.[16] The variable results from trials summarized in Table 1 have contributed to the lack of consensus regarding "standard" chemotherapy for patients with advanced HCC; they have also resulted in ongoing debate regarding the best control arm for future randomized trials.

Furthermore, the definition of *drug activity* has changed over the years. It is now well recognized that the conventional markers of radiographic response (World Health Organization [WHO] or RECIST criteria) are poorly related to tumor cell kill in liver tumors and that end points other than radiographic tumor shrinkage, such as time to tumor progression, progression-free survival, and certainly overall survival, are more meaningful measures of therapeutic benefit.[17]

The third set of reasons is related to HCC tumor biology and intrinsic or acquired drug resistance. Large HCCs commonly develop areas of central necrosis, which may inhibit drug delivery to actively growing parts of the tumor. Topoisomerase IIa encodes an enzyme that is the target for anticancer chemotherapeutic agents such as doxorubicin, and mutations are associated with resistance.[18] There is upregulation of topoisomerase IIa in doxorubicin-resistant HCC cell lines, and its expression is associated with an aggressive tumor phenotype.[19] Cancer cells, including HCC cells, often have intrinsic drug resistance mediated by enhanced cellular drug efflux of several cytotoxic agents. This phenomenon is associated with increase in a drug transporter family, the adenosine triphosphate–binding cassette proteins that include MDR1, p-glycoprotein (p-gp), and the multidrug resistance protein (MRP).[20,21] Both these are overexpressed in HCC.[21,22] Overexpression of MDR1 accompanied by a decrease in doxorubicin accumulation levels has been observed in certain HCC cell lines.[23] The *H19* gene is believed to induce p-gp expression and MDR1-associated drug resistance in HCC cells through regulation of MDR1 promoter methylation.[23] Coexpression of p53 and p-gp may contribute to HCC drug resistance in HCC cell lines.[24] In addition, recent evidence suggests that hypoxia, MDR1 expression, and an angiogenic HCC phenotype may be linked.[25,26]

Clearly, to improve the outcome for patients with advanced HCC, alternatives to traditional cytotoxic chemotherapy agents must be explored.

HCC IN THE ERA OF TARGETED THERAPIES

In recent years, several molecular targets, including oncogenes, oncoproteins, and cellular receptors, have been identified in a variety of cancers as being key elements in carcinogenic pathways. Consequently, several agents have been developed that, by a variety of mechanisms, interfere with cell signaling and have demonstrated anticancer activity. In some cancers, the molecular target–targeted agent relationship is well understood—for example, the monoclonal antibody trastuzumab is only effective in tumors in which the her-2/neu oncoprotein is amplified. Conversely, there are several agents that target the transmembrane epidermal growth factor receptor (EGFR) and have demonstrated survival benefit in a broad range of tumor types, yet little is understood regarding the relationship between target expression and agent efficacy or lack thereof. Several targeted or novel biologic agents are now being tested in HCC patients. This discussion focuses on those aspects of hepatocarcinogenesis that are sufficiently well understood to provide a rational basis for developing clinical trials that use existing novel or targeted therapies in HCC.

TARGETING CARCINOGENIC PATHWAYS IN HCC

Hepatocarcinogenesis is known to be a complex multistep process that results in a large number of heterogeneous molecular abnormalities, and thus numerous potential targets for existing therapeutic agents. The pathways summarized below represent rational targets for existing therapeutic agents in HCC.

Mitogen-Activated Protein Kinase Pathway

The mitogen-activated protein kinase pathway (MAPK) pathway involves a cascade of phosphorylation of four major cellular kinases; ras, raf, MAP, and ERK (MAP, mitogen-activated protein kinase; ERK, extracellular-signal-regulated kinase), which is responsible for cellular proliferation and differentiation. These intermediates are found to be high in both HCC cell lines and human specimens.[27–31] Therapeutic agents that target this pathway include sorafenib (targets both raf and vascular endothelial growth factor receptor, VEGFR) and farnesyl transferase inhibitors (targeting ras). A phase 2 trial of sorafenib demonstrated antitumor activity in advanced HCC patients. This study did not meet its primary end point of response on the basis of WHO criteria, with limited response rate of 2.2%. However, many patients (33.6%) had stable disease for at least 4 months, with many showing central tumor necrosis.[32] On the basis of the encouraging overall survival of 9.2 months reported in the phase 2 trial, a placebo-controlled international trial was conducted in HCC patients with Childs-Pugh A cirrhosis. Preliminary data presented in abstract form from the phase 3 trial showed better survival in the sorafenib arm (10.7 months) compared with placebo (7.9 months).[33] These results indicate that this agent offers a survival advantage compared with placebo and with several cytotoxic agents (based on historical controls), but this may be comparable to survival observed with other biologic agents (Table 1).

PI3K/AKT/mTOR Pathway (Phosphoinositide-3 Kinase/Protein Kinase B/Mammalian Target of Rapamycin)

This kinase cascade is responsible for cellular proliferation and apoptosis, and is closely linked to cell cycle. PI3K is associated with cell surface growth factor receptors, and on ligand binding can trigger formation of PIP3, which in turn activates Akt and leads to a number of downstream events (mTOR being one of the targets). This pathway is known to be upregulated in a subset of HCC patients.[34–36] Molecular target therapy such as rapamycin, a naturally occurring mTOR inhibitor, showed promising results in HCC cell lines,[37,38] but no results from clinical trials of any agents that target mTOR in HCC patients are available.

Epigenetic Changes

Epigenetic modifications of the genome (mainly hypermethylation of CpG island and histone deacetylation) are accumulated during hepatocarcinogenesis in chronically injured liver. A large number of tumor suppressor genes have been shown to be inactivated by epigenetic mechanisms in HCC. Success in epigenetic therapy (such as 5-aza-2′-deoxycytidine and SAHA) had been achieved in both hematological malignancies and solid tumors. In HCC cell lines, chemosensitivity can be potentiated by epigenetic therapy.[39,40] A multicenter phase 1/2 trial on a novel histone deacetylase inhibitor, PXD-101, is currently underway in Hong Kong.

GROWTH FACTORS AS THERAPEUTIC TARGETS IN HCC

The epidermal growth factor receptor (EGFR) is frequently expressed in human hepatoma cells, and EGF may be one of the mitogens needed for the growth of hepatoma cells.[41,42] Several agents that inhibit EGF signaling are clinically available, including gefitinib, cetuximab, erlotinib, and panitumumab. Erlotinib is an orally active and selective inhibitor of the EGFR/HER1-related tyrosine kinase enzyme. EGFR/HER1 expression was detected in 88% of the patients in a phase 2 study of erlotinib.[43] In two phase 2 studies of this agent, the response rates were was < 10%, but the disease control rate was > 50%, and median survival times were 10.75 and 13 months, respectively.[43,44] Other studies of anti-EGFR agents in HCC are summarized in Table 1.

HCCs are generally hypervascular, and vascular endothelial growth factor (VEGF) promotes HCC development and metastasis.[45–49] Various agents targeting the VEGF circulating ligand or transmembrane receptor, including bevacizumab (Avastin), sorafenib (Nexavar), and TSU-68, have been studied in patients with HCC. Bevacizumab, a monoclonal antibody inhibitor of VEGF ligand, has been investigated in phase 2 studies alone or combination with other agents. These studies showed a high disease control rate of > 80% and a median progression-free survival of > 6 months.[50] Sorafenib, an oral multikinase inhibitor, blocks tumor cell proliferation mainly by targeting Raf/MEK/ERK signaling at the level of Raf kinase, and exerts an antiangiogenic effect by targeting VEGFR-2/-3.[51–54] TSU-68 is an oral antiangiogenesis compound that blocks VEGFR-2 (vascular endothelial growth factor receptor), PDGFR (platelet-derived growth factor receptor), and FGFR (fibroblast growth factor receptor); a phase 1/2 study has been conducted in Japan.[55]

IMMUNOTHERAPY OF HCC

Increasing evidence suggests that immune responses are important in the control of cancer and that the manipulation of the immune system to recognize and attack tumors may be a valuable form of therapy. HCC is attractive target for immunotherapy, because in addition to documented cases of spontaneous regression, lymphocytes can be seen infiltrating tumors and tumor-associated proteins such as alfafetoprotein (AFP) could act as targets for immune-mediated attack.[56,57] Given the limitations of current treatment modalities in the treatment of HCC, interest has been stimulated in immunotherapy of HCC, and a number of promising strategies have been developed in the laboratory, some of which have been applied in the clinical setting.

HCC are often infiltrated with lymphocytes, and patients with higher levels of tumor-infiltrating lymphocytes have a better prognosis after resection and transplantation.[56] A randomized, controlled clinical trial has shown that disease-free survival after HCC resection can be increased by infusion of lymphocytes activated by anti-CD3 and interleukin 2, suggesting a promising role for T cell adoptive immunotherapy.[58]

To generate a tumor-specific immune response, tumor-associated antigens must be presented to the immune system in an immunostimulatory context. Dendritic cells (DC) are the most efficient method of stimulating immune responses and are potent inducers of antitumor immunity when loaded with tumor-associated antigens. Animal models have shown encouraging results for DC-based vaccination strategies. DC transduced with adenovirus encoding AFP were able to prevent or delay growth of an AFP-producing tumor cell line in mice, and this was accompanied by the appearance of AFP-specific cytotoxic T lymphocytes.[59] By using fusions of DC and syngeneic hepatoma cells, Kawada and colleagues[60] were able to prevent the growth of implanted hepatoma cells and prevent local recurrence after surgical resection in rats.

The success of animal models in DC-based immunotherapy of HCC has lead to a number of clinical studies. These studies are all small and are mainly phase 1/2 studies designed primarily to assess feasibility and tolerability of this treatment modality. Currently reported DC vaccination studies have used DC loaded with autologous tumor or hepatoma cell line lysates.[61] DC have also been directly injected into tumors.[62] Clinical responses to these approaches have at best been modest, and the success of animal vaccination studies has not to date been replicated.

There are many reasons why this should be. First, the patients selected for clinical studies have been those with advanced disease and therefore may have tumor-induced immunosuppression. Additionally, questions still remain about the optimal route of administration of DC vaccines and the optimal method of loading tumor-associated antigens needs to be established. Importantly, the effect of concomitant viral infection, especially with hepatitis C virus, needs to be clarified.[63]

Currently the role for immunotherapy in HCC is limited, but from studies performed so far, we can be certain that future clinical studies should be ran-

domized and include patients with earlier disease and small tumor burden, to better identify potential benefit and truly identify the role of immunotherapy in HCC.

CLINICAL TRIAL DESIGN FOR BIOLOGIC AGENTS IN HCC

As noted previously, the availability in the clinic of several novel biologic agents and the urgent need for effective therapies for advanced HCC has led to the evaluation of many of these agents in HCC, principally in phase 2 trials. The SHARP trial[33] was the first to demonstrate a statistically significant survival benefit for any chemotherapy agent in patients with HCC. This trial was, however, conducted in patients with excellent performance status and well-preserved liver function. The efficacy and safety of sorafenib in patients with more tumor-related symptoms and advanced hepatic dysfunction remains to be established.

A key objective going forward is to assess new agents and to integrate these and sorafenib into the treatment of all stages of HCC and patients with Child-Pugh A and B cirrhosis. The classic approach is to evaluate new agents in single-arm phase 2 studies and use classic radiological response criteria such as WHO or RECIST as a measure of activity and thereby identify promising agents to take forward into phase 3 clinical trial testing against an appropriate control group. This approach, however, is being questioned because traditional radiographic tumor responses may not occur with biologic agents, although they may cause other anticancer effects that may lead to meaningful patient benefit. This is especially true in HCC, where radiological assessment is notoriously difficult because of poor delineation of tumors in the liver[64] and tumor necrosis may occur without any change in overall tumor dimensions.

These observations have led some investigators to develop phase 2 studies with a major focus on correlative studies that may help delineate a mechanism of action for a particular drug (e.g., a kinase inhibitor along one of the different cell cycle pathways) such as downregulation of a downstream kinase which may predict response,[32] or by the use of novel radiological techniques that use changes in blood flow as criteria by which to assess biologic activity of antiangiogenic therapies.[32] Another option is to use the randomized phase 2 trial design that, by providing a contemporary control group, may permit a more confident assessment of the likelihood that a particular agent is worthy to progress to phase 3 trials.[65]

CONCLUSIONS

Conducting controlled clinical trials of systemic chemotherapy regimens in HCC patients is challenging. Obstacles include the multiple comorbidities of patients with cirrhosis, the intrinsic chemoresistance of HCC, the advanced nature of HCC at the time of presentation in most patients, the pharmacotherapeutic challenges of treating a cancer that arises in an already damaged liver, and the distribution of patients primarily in developing nations where multidisciplinary treatment of HCC may not be available. HCC is a heterogeneous disease in terms of its cause, underlying associations, and biologic and clinical behavior, which further complicates clinical trial design. The need for effective systemic therapies for HCC patients is clearly evident, and making progress in this area requires the talent and expertise of all of the medical disciplines involved in the care of HCC patients.

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Critical Reviews in Oncology/Hematology 67 (2008) 8–15

CRITICAL REVIEWS IN
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Growth factors as therapeutic targets in HCC

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Accepted 26 February 2008

Contents

Abstract

Despite various effective local treatments for hepatocellular carcinoma (HCC), some patients do not meet the treatment criteria because of extrahepatic metastases or macroscopic vascular invasion at the time of their diagnosis. Furthermore, many patients treated with successful local treatments develop recurrences after treatment. Although these patients receive systemic treatment including chemotherapy, HCC is generally recognized as a chemo-resistant tumor. Recently, new molecular targets have been confirmed and various targeted agents are now being investigated for the treatment of HCC. Epidermal growth factor receptor (EGFR) is frequently expressed in human hepatoma cells, and EGF may be one of the mitogens that are needed for the growth of hepatoma cells. HCC is generally hypervascular, and vascular endothelial growth factor (VEGF) promotes HCC development and metastasis. Various inhibitors targeting EGFR and/or VEGF, VEGF receptor (VEGFR) have been developed as treatments of HCC. In phase-II studies of these growth factor inhibitors, the response rates are relatively low; however, high rates of disease control, enabling a good time to progression, have been achieved. Recently, a randomized phase III trial of sorafenib versus placebo conducted in patients with advanced HCC demonstrated the beneficial effects of this drug on the time-to-progression and overall survival of the patients, and the drug could become established as the standard chemotherapeutic agent for advanced HCC. Further clinical trials using biologic agents are warranted to prolong the survival in HCC patients.

Keywords: Hepatocellular carcinoma; Epidermal growth factor receptor; Vascular endothelial growth factor

1. Introduction—etiology and treatment strategy

Hepatocellular carcinoma (HCC) is the fifth most common malignancy worldwide [1], with approximately 500,000 new cases per year. There are globally some discrepancies in the incidence and etiology of HCC. Almost 80% of patients with HCC arise in Asia and Africa [2]. In Japan, approximately 34,000 patients died of primary liver cancer, and it is 11.6% of cancer deaths and the fourth mortality of malignancy [3]. The incidence of HCC is approximately 43/100,000 population, and about 50,000 new HCC patients annually arise [3]. Most patients with HCC have chronic liver disease, especially liver cirrhosis, and it is mainly due to hepatitis virus infection. However, there is definitely difference in etiology among regions. Hepatitis B virus (HBV) is very common in east and South-East Asia and Africa; more than 80% HCC patients have HBV infection [2]. On the other hand, hepatitis C virus

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doi:10.1016/j.critrevonc.2008.02.009

(HCV) is common in Japan, and HCV antibody is observed in 72% of Japanese HCC patients [4]. The incidence of HCV infection is also increasing in the United States and European countries, and the incidence of HCC is rising [5].

The treatments are classified to local and systemic therapy. Various treatment modalities such as surgery, ablation, transcatheter arterial chemoembolization (TACE), and liver transplantation, are available as local therapeutic approach. The treatments for HCC are selected according to the tumor stage, the grade of liver dysfunction, and performance status [4,6]. The local approaches yield good outcomes in patients with earlier stage disease. Since rigorous screening of at-risk patients is carried out and many early-stage HCC patients are identified, large numbers of patients probably undergo surgery or regional therapy in Japan. In other parts of the world, the majority of HCC patients may have advanced disease at the time of diagnosis.

Despite successful these local therapies above, many patients develop recurrences or progression after treatments. Some patients do not meet the indication criteria of local therapies at the time of their diagnosis, such as extrahepatic metastases. Although these patients receive systemic treatment including chemotherapy, HCC is generally recognized as a chemo-resistant tumor. Recently, some growth factors and various signal transduction pathways have been identified, and various targeted agents are now being investigated as the treatment of HCC. They control processes of cell proliferation and survival and specialized functions such as angiogenesis. Dysregulated signaling pathways contribute to malignant transformation in human cells. In this paper, recent progress in treatments using growth factor targeted agents for HCC is reviewed.

2. Systemic chemotherapy for hepatocellular carcinoma

Systemic chemotherapy is applied for patients with advanced HCC to which local treatments are not able to be indicated. TACE refractory stage is also considered candidates for chemotherapy. In various reports on chemotherapy for HCC, anthracycline antitumor antibiotic agents such as doxorubicin and mitoxantrone have been considered as the basis of chemotherapy [7,8]. Furthermore, cisplatin and/or fluorouracil have been used as combination chemotherapy [9–12]. The response rate ranges from 14% to 26%, and the median overall survival (OS) varies from 8.9 to 11.6 months in combination chemotherapies of fluorouracil/mitoxantrone/cisplatin (FMP), epirubicine/cisplatin/fluorouracil (ECF), and cisplatin/interferon α-2b/doxorubicin/fluorouracil (PIAF). Doxorubicin has been considered a referential arm in randomized clinical trials for HCC based on comparison trial between doxorubicin and supportive treatment [9]. Despite better response in phase III trials of combination chemotherapy compared to doxorubicin, no standard chemotherapy has currently been identified that can clearly prolong survival; for example, recent phase III trial of doxorubicin versus PIAF failed to show survival benefit (response rate: 10.5% for doxorubicin and 20.9% for PIAF, $p = 0.058$; median OS: 6.8 months for doxorubicin and 8.7 months for PIAF, $p = 0.83$) [13].

In Japan, various regimens of hepatic arterial infusion chemotherapy have been tried for very advanced stage HCC such as extensive portal vein tumor thrombus, and some regimens were reported to have response rate of more than 40% [14,15]. However, no standard regimen has currently been identified that can clearly prolong survival based on prospective large clinical trials.

1.1. Epidermal growth factor

Cell regulation is controlled by secreted polypeptide molecules called growth factors such as epidermal growth factor (EGF) and transforming growth factor (TGF)-α. There are four different human EGF receptors, human epidermal growth factor receptor (HER) 1–4, and EGF receptor exists as monomers and consists of extracellular domain and intracellular domain. When EGF binds to EGF receptor, a dimerization loop of EGF receptors is induced at first. Then, tyrosine-kinase intracellular domain is activated and it serves as docking sites for intracellular signaling molecules that bind to phosphotyrosine. It leads various pathways to cancer cell proliferation, invasion, metastasis, angiogenesis, and inhibition of apoptosis [16,17]. EGFR/HER1 is frequently expressed in human hepatoma cells or HCC, and EGF may be one of the mitogens that are needed for the growth of hepatoma cells [18–21]. The ligand for EGFR/HER1 has different effects on different human hepatoma cell lines, and its role might be more important in poorly differentiated hepatoma cells than in the well-differentiated ones [18]. Increased expression of TGF-a and EGFR also occur frequently in human HCC, and the detection of greater staining in more highly differentiated portions of the tumors suggested that increased expression of TGF-α and EGFR/HER1 might be related to the early stages of human hepatocarcinogenesis [20]. Thus, in patients with HCC, the EGFR/HER1 blockade possible reduces HCC development and/or delays the disease progression.

In cellular signalings, various sites of EGF receptor can be targets of treatments for cancer. Regarding development of agents inhibiting the tyrosine kinase activity, there are a variety of strategies targeting both the extracellular and intracellular domains [22]. Small molecule compounds, which directly inhibit tyrosine phosphorylation, have been investigated for HCC. So far, erlotinib and lapatinib have been reported in clinical trials for HCC (Table 1). Overexpression of EGFR/HER1 was observed in 52–71% of the patients with HCC in phase-II studies of erlotinib or cetuximab [23,24,27]. The response rate to erlotinib was relatively low, but the DCR was significant (43–59%), and the 6-month progression-free survival rate was 28–32% [23,24]. In these trials, the disease control rate (DCR), defined as the sum of the CR + PR + SD

Table 1
Clinical trials of epidermal growth factor receptor targeted agents for hepatocellular carcinoma

	Class of anti-EGF agent	Targets	Study design	*n*	Response rate (%)	Disease control rate (%)	PFS at 6 months (%)	Median PFS	Median OS (mo)	Author	Year
Erlotinib	Tyrosine-kinase inhibitor	HER1/EGFR	Phase II	38	9	59	32	3.2 mo	13.0	Philip	2005 [23]
Erlotinib			Phase II	40	0	43	28	3.1 mo	6.3	Thomas	2007 [24]
Lapatinib		HER1/EGFR + HER2	Phase II	40	5	38	–	2.3 mo	6.2	Ramanathan	2006 [25]
Cetuximab	Antibody	HER1/EGFR	Phase II	32	0	44	–	8 wk	–	Gruenwald	2007 [26]
Cetuximab			Phase II	30	0	17	3	1.4 mo	9.6	Zhu	2007 [27]
GEMOX/cetuximab			Phase II	45	24	65	42	4.5 mo	9.2	Louafi	2007 [28]

Response = complete response + partial response; disease control = complete response + partial response + stable disease; PFS, progression-free survival; OS, overall survival.

rates, was evaluated. The antitumor activity was basically evaluated based on the Response Evaluation Criteria in Solid Tumors (RECIST), and CR or PR needed to be confirmed at least 4-week intervals, and the SD, at 6–8-week intervals. Evaluation by CT or MRI was performed every 6–8 weeks. Although the interval for assessment of the CR, PR and SD has been shown to vary among trials, the DCR appears to vary little, and it seems evaluation of the DCR may be a valid approach in these trials. In those phase II trials of tyrosine-kinase inhibitors in HCC, the response rate and the median progression-free survival (PFS) or time-to progression (TTP) were similar, from 0% to 9% and from 2 to 3 months, respectively, whereas the median OS varies from 6 to 13 months (Table 1). Although both erlotinib and lapatinib administered alone were well tolerated (Table 3), their antitumor activity against HCC seems modest.

Cetuximab is a chimeric monoclonal Ig G1 antibody directed against the EGFR and blocks binding of endogenous EGFR ligands (particularly of EGF and TGF-α). Cetuximab was investigated whether it have the potency of anti-neoplastic effect in human HCC cells [29]. The study showed that cetuximab inhibited growth of p53 wild-type HepG2 HCC cells. Furthermore, the combination of cetuximab with tyrosine-kinase inhibitors, the HMG-CoA-reductase inhibitor fluvastatin or doxorubicin resulted in synergistic antiproliferative effects [29]. In 2007, three phase-II studies using cetuximab have been reported (Table 1). Although cetuximab administered alone seemed to be well tolerated (Table 3), the drug demonstrated little antitumor activity against HCC. Cetuximab alone or combination of cetuximab and cytotoxic chemotherapy appears not to be worthy of further investigation of a large-scale phase III trial.

1.2. Vascular endothelial growth factor

HCC is generally hypervascular and contains rich tumor vasculality. Vascular endothelial growth factor (VEGF) is related to angiogenesis and one of the important factors involved in the angiogenesis of HCC [30–33]. Moreover, VEGF promotes HCC development and metastasis, and serum VEGF level is a significant independent prognostic factor in patients with HCC [32,34]. The mechanism of effects of anti-VEGF agent has been examined. Solid tumors including HCC require blood vessels for growth. Tumor vessels are structurally and functionally abnormal, contributing to the increase in interstitial fluid pressure within the tumor [35–37]. Anti-VEGF treatment leads to pruning of the tumor vasculature, reduction in vessel tortuosity, and a drop in interstitial fluid pressure, a process termed vessel normalization [35–37]. Furthermore, it was reported that combination of a cytotoxic drug with anti-VEGF agent leads to a rapid decrease in interstitial fluid pressure, which may enhance the delivery of chemotherapy to tumor cells [37]. As a result, tumor reduction and improvement of survival rates are led. Various inhibitors targeting VEGF have been developed for the treatment of HCC. Some VEGF or VEGF receptor (VEGFR)

targeting agents have been investigated for HCC treatments in phase I and/or II studies (Table 2).

Bevacizumab (Avastin; Genentech Inc., South San Francisco, CA), a recombinant, humanized monoclonal antibody that targets VEGF [37], has been investigated in phase-II studies of bevacizumab alone or combination with other agents [38–42]. These studies showed that response rates were moderate from 10% to 20% and achieved a high disease control rate of 47–90%. The median PFS was also achieved very promising, ranging from 5.3 to 9.0 months, especially combination of bevacizumab and erlotinib [41]. Further large-scale clinical trials of the combination therapy, such as bevacizumab and erlotinib, are warranted.

Various small molecular multi-kinase inhibitors including VEGFR have been investigated for HCC. Sorafenib, an oral multi-kinase inhibitor, blocks tumor cell proliferation mainly by targeting Raf/MAPK-ERK kinase (MEK)/extracellular signal regulated kinase (ERK) signaling at the level of Raf kinase, and exerts an antiangiogenic effect by targeting VEGFR-2/-3 [43]. Addition a role of VEGFR for development and progression of HCC, some studies demonstrated that MAPK/ERK activation in human HCC play an important role in multistep hepatocarcinogenesis, especially in the progression of HCC [48,49]. Over-expression of activated MEK1in HCC cell lines enhanced tumor growth and survival by preventing apoptosis [50]. Therefore, blocking MEK-MAPK activity through Raf kinase may offer therapeutic benefits in HCC. In a phase II study of sorafenib, the response rate was only 2%, but 4.2 months of the median TTP was achieved [43]. A phase I study of sorafenib in Japanese HCC patients showed that the safety profile was similar to that in the phase II study of sorafenib in the USA and that the recommended dose was the same, dose of 400 mg bid in the USA. The response rate was 4%, and the median TTP and the OS were 4.9 and 15.6 months, respectively, in Japanese HCC patients [44]. TSU-68 is an oral anti-angiogenesis compound that blocks VEGFR-2, PDGFR, and FGFR. The phase I/II study showed that it was well tolerated in patients with HCC and that the response rate was 7%. The study was reported on going at that point [45]. Sunitinib is also an oral multitargeted receptor tyrosine kinase inhibitor with active against VEGFR, PDGFR, and c-kit. It was reported that sunitinib has also activity against advanced HCC in the phase II study [46,47].

At the 2007 ASCO annual meeting, a randomized phase III trial of sorafenib versus placebo in patients with advanced HCC was reported [51]. Six hundreds and two patients were randomized to two groups. The time-to progression was 5.5 months for sorafenib and 2.8 months for placebo, and the hazard ratio for sorafenib was 0.58 (95% CI: 0.44–0.74; $p = 0.000007$). The median OS was 10.7 months for sorafenib and 7.9 months for placebo, and the hazard ratio for OS for sorafenib was 0.69 (95% CI: 0.55–0.87; $p = 0.00058$). This trial demonstrated a statistically significant improvement not only in the progression but also in the survival by sorafenib in those patients. Thus, sorafenib is the first systemic therapy

Table 2
Clinical trials of vascular epidermal growth factor (VEGF) or VEGF receptor targeted agents for hepatocellular carcinoma

	Class of anti-VEGF agent	Targets	Study design	*n*	Response rate (%)	Disease control rate (%)	PFS at 6 months (%)	Median PFS (months)	Median OS (months)	Author	Year
Bevacizumab			Phase II	30	10	80	–	6.5	–	Schwartz	2006 [38]
Bevacizumab			Phase II	30	13	67	–	–	–	Malka	2007 [39]
GEMOX/bevacizumab	Antibody	VEGF	Phase II	33	20	47	48	5.3	9.6	Zhu	2006 [40]
Bevacizumab/erlotinib			Phase II	34	21	79	–	9.0	19.0	Thomas	2007 [41]
Cape/Ox/bevacizumab			Phase II	32	13	90	–	4.5	10.6	Sun	2007 [42]
Sorafenib		Raf/VEGFR-	Phase II	137	2	42	37	4.2	9.2	Abou-Alfa	2006 [43]
Sorafenib	Small-molecule inhibitor	2,3/PDGFR	Phase 1	25	4	80	–	4.9	15.6	Furuse	2008 [44]
TSU-68		VEGFR-2, PDGFR, FGFR	Phase I/II	15	7	47	–	–	–	Kanai	2006 [45]
Sunitinib		VEGFR-2, PDGFR, c-Kit	Phase II	26	4	42	–	4.1	11.6	Zhu	2007 [46]
			Phase II	37	3	38	–	4.8	10.4	Faivre	2007 [47]

GEMOX, combination regimen of gemcitabine and oxaliplatin; cape, capecitabine; ox, oxaliplatin, response = complete response + partial response; disease control = complete response + partial response + stable disease; PFS, progression-free survival; OS, overall survival.

Table 3
Major toxicity of growth factor inhibitors

Agent	Major toxicity	Author	Year
Erlotinib	Rash, fatigue, diarrhea, bilirubin elevation, anorexia, nausea, vomiting	Philip	2005 [23]
Erlotinib	Diarrhea, rash, fatigue, nausea, pruritus	Thomas	2007 [24]
Lapatinib	Elevation of liver enzymes, diarrhea, nausea, vomiting, rash, anemia, thrombocytopenia	Ramanathan	2006 [25]
Cetuximab	–	Gruenwald	2007 [26]
Cetuximab	Acneiform rash, fatigue, hypomagnesemia, nausea	Zhu	2007 [27]
GEMOX/cetuximab	Thrombocytopenia, neutropenia, acneiform rash, asthenia, neurotoxicity	Louafi	2007 [28]
Bevacizumab	Transient ischemic attack, fatigue, abdominal pain, gastric ulcer, hypertension, hyperbilirubinemia, rash, proteinuria	Schwartz	2006 [38]
Bevacizumab	Transient ischemic attack, hemorrhagic ascites, proteinuria, fatigue, epistaxis, hypertension, proteinuria	Malka	2007 [39]
GEMOX/bevacizumab	Hypertension, proteinuria, epistaxis, hematochezia, upper GI bleed, small bowel perforation	Zhu	2006 [40]
Bevacizumab/erlotinib	AST/ALT elevation, hyperkalemia, acne, diarrhea, proteinuria, GI bleed, fatigue, hypertension	Thomas	2007 [41]
Cape/Ox/bevacizumab	Peripheral neuropathy, fatigue, hand–foot syndrome, gastrointestinal perforation, sepsis, dizziness, neutropenia	Sun	2007 [42]
Sorafenib	Hand–foot skin reaction, rash, fatigue, diarrhea	Abou-Alfa	2006 [43]
Sorafenib	Lymphopenia, thrombcytopenia, weight loss, hand–foot skin reaction, rash, pruritus, diarrhea, elevation of lipase and amylase	Furuse	2008 [44]
TSU-68	Hypoalbuminemia, diarrhea, abdominal pain, fever, AST/ALT elevation	Kanai	2006 [45]
Sunitinib	Neutropenia, lymphopenia, AST/ALT elevation, fatigue, rash, thrombocytopenia	Zhu	2007 [46]
	Thrombocytopethrombocytopenia, neutropenia, AST elevation, CNS symptoms, asthenia, hemorrhage	Faivre	2007 [47]

GEMOX, combination regimen of gemcitabine and oxaliplatin; cape, capecitabine; ox, oxaliplatin.

to prolong survival in advanced HCC patients. In addition to using sorafenib, investigation of new chemotherapeutic regimens in large-scale RCTs using sorafenib-alone as the control treatment is proposed. Furthermore, the suitability of using sorafenib in various other settings besides the advanced HCC setting, such as in the adjuvant setting and in combination with other agents is also expected to be examined.

1.3. Future directions in relation to the use of growth factor inhibitors

Based on the results of clinical trials using EGF and VEGF receptor inhibitors, the possibility of synergetic effects between the biologic agents and cytotoxic therapies should be discussed. Although gemcitabine + oxaliplatin (GEMOX) and capecitabine + oxaliplatin (Capox) in combination with bevacizumab have been investigated as regimens combining cytotoxic agents and a biologic agent, these combination regimens did not seem to be superior to bevacizumab alone [38,39,42,43]. While the response rate and PFS in patients treated with the combination of GEMOX and cetuximab appeared to be better than those in the patients treated with cetuximab alone, the survival benefit was not clear [26–28]. On the other hand, combinations of EGFR and VEGF inhibitors such as erlotinib and bevacizumab have shown very promising results [42]. So far, combinations of cytotoxic chemotherapy + biologic agents or of two or more biologic agents have not been sufficiently investigated. Therefore, the usefulness of these combinations must be investigated.

Most molecular targeted agents including growth factor inhibitors are regarded as cytostatic agents. While the survival benefit should be finally evaluated in a phase III study because of the heterogeneity of HCC, various endpoints as surrogate markers are selected in phase-II studies. Although response rate is often set as primary endpoint in small phase-II studies of cytotoxic chemotherapy, the appropriate endpoints should be considered in phase-II studies of molecular targeted agents, especially for HCC that is known as chemo-resistant. In two sorafenib studies in the USA and Japan, the response rate was only 2–4% and the disease control rate varies 42–80%, whereas the TTP was approximately equal, 4.2 and 4.9 months. In a phase III study of sorafenib, the TTP was also 5.5 months, similar to that in phase-II studies, while that of placebo arm was 2.8 months [51].

The existence of a relationship between the TTP/PFS and OS has been reported in patients with other solid tumors [52,53]. A correlation between PFS and OS was reported in colorectal cancer patients receiving chemotherapy with 5-FU/leucovorin and bevacizumab [52]. The TTP was also shown to be well correlated to the OS in a

pooled analysis of many phase-II studies for biliary tract cancer [53]. On the other hand, a trend for marked variability of the OS as compared with that of the TTP/PFS has been observed in phase-II studies for HCC, which could probably be ascribed to the heterogeneity of HCC. Thus, assessment of the TTP is probably more suitable for assessing the efficacy in phase-II studies for HCC. Furthermore, investigation of the possible existence of a correlation between TTP/PFS and OS is required in patients with HCC who are treated with growth factor inhibitors, in order to establish an appropriate design of phase-II studies for HCC.

Regarding indication of molecular targeted therapy, there are two important targets of HCC stages. One is the treatment for advanced HCC like conventional chemotherapy to prolong the survival in patients with metastatic disease or TACE refractory disease who cannot be treated with local treatments. The other is an adjuvant treatment after local treatments like surgical resection, ablation therapy, and TACE. Rather than gross advanced tumor, tiny residual tumors after these local treatments seem to be effectively treated with cytostatic agents like growth factor inhibitors. Furthermore, there is possibility to prevent promotion to cancer from precancerous lesion like dysplastic nodules accompany with cirrhotic liver using growth factor inhibitors. In the future, In addition to the assessment of its usefulness for advanced-stage HCC, investigation of the usefulness of molecule-targeted therapy in the adjuvant or prophylactic setting is also expected.

2. Conclusions

Despite multidiscipline treatments for HCC effective systemic therapies is necessary to improve the survival of HCC patients. Numerous growth factor inhibitors such as gefitinib, erlotinib, cetuximab, trastuzumab, bevacizumab, sunitinib and sorafenib have recently approved in a variety of solid tumors based on results of RCTs. In patients with advanced HCC, the survival benefit of sorafenib has been demonstrated in a RCT for the first time, and the drug has recently been approved for the treatment of HCC in Europe and USA. Moreover, other promising agents also need to be developed for prolonging the survival further in HCC patients. Sunitinib has shown promising activity against HCC in phase-II studies, and large-scale randomized trial of sunitinib is warranted. Some agents targeted selectively at VEGFR are currently being investigated for various solid tumors, and they may eventually also come to be applied for the treatment of HCC.

Although further conducting clinical trials of systemic chemotherapy including growth factor inhibitors is required, there are various obstacles such as heterogeneity of HCC and underlying chronic liver disease. Since HCC is a heterogeneous disease in terms of its etiology and various degrees of underlying chronic liver disease, preliminary clinical trials like phase I study may need to investigate the pharmacokinetics or optimal dose of chemotherapeutic agents. Liver dysfunction due to chronic liver disease often affects the efficacy of chemotherapy. Therefore, the efficacy needs to be evaluated according to the degree of impairment of liver function (Child-Pugh classification).

Furthermore, numerous factors which affect the prognosis in patients with HCC including the stage, the degree of liver damage and performance status require well-designed clinical trials. In addition, to establish optimal chemotherapy using growth factor inhibitors, identification of molecular profiles using tissue specimens and proper patient selection may be necessary.

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Biography

Junji Furuse currently works as a medical doctor at Division of Hepatobiliary and Pancreatic Oncology, National Cancer Center Hospital East, Japan. In 1984, he graduated from Chiba University of Medical School and then attended the First Internal Medicine Department in Chiba University. After completing residency in internal medicine, he specialized in gastroenterology and hepatology between 1988 and 1992. Since 1992, he has worked as a medical oncologist at the National Cancer Center Hospital East. He has just launched division of medical oncology as a professor in School of Medicine, Kyorin University in March 2008. Dr. Furuse is focusing his clinical practice and research in the areas of medical treatment of hepatobiliary and pancreatic cancers. He is the principal investigator on several clinical trials of systemic chemotherapy and/or chemoradiotherapy in hepatobiliary and pancreatic cancer, and a co-investigator on numerous clinical trials. He has contributed to the writing of over 200 articles and abstracts, some of which he has also presented at various international and national congresses. Dr. Furuse is an active member of many societies not only in Japan also internationally including the American Society of Clinical Oncology and the European Society of Medical Oncology.

2008年5月号(Vol. 31 No. 6)

トピックス

分子標的治療

Targeted therapy for hepatocellular carcinoma

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●要旨●肝細胞癌における治療戦略上，これまで化学療法はほとんど考慮されてこなかった。上皮成長因子などの細胞シグナル伝達経路や血管内皮増殖因子など，癌の増殖，転移に関する分子生物学的特性に基づく分子標的薬の開発が進み，化学療法の環境も変化してきた。肝細胞癌では従来の殺細胞性抗癌剤は併用療法により高い奏効率が得られるものの，生存期間の延長にはつながらず，標準的化学療法は確立していなかった。2007年，マルチキナーゼ阻害薬sorafenibは無作為化比較試験により奏効率は2～4％と低いものの，placebo群に比べ無増悪生存期間と全生存期間の延長が報告された。sorafenibは肝細胞癌の標準的化学療法と位置付けられるものと期待されている。さらに今後，新しい分子標的薬や併用療法などの試み，局所治療の補助療法など肝細胞癌の治療戦略に大きな変化がもたらされるものと考えられる。

● key words：肝細胞癌，上皮成長因子受容体，血管内皮細胞増殖因子受容体，全身化学療法

はじめに

肝細胞癌の治療は一般に癌進行度と肝障害度に応じて治療選択が行われ，肝切除などの局所療法から化学療法までその治療法は多岐にわたる。肝細胞癌に対する治療選択については日本では『肝癌診療ガイドライン』による肝細胞癌治療アルゴリズムが公表されている[1]。またBarcelona groupによるBarcelona Clinic Liver Cancer（BCLC）staging classification[2]は進行度と全身状態（performance status；PS）や肝障害度から治療選択を示し，ヨーロッパを中心に用いられている。これらの治療選択のガイドラインにおいて，肝切除やラジオ波（RFA）など局所壊死療法，肝移植，肝動脈塞栓化学療法（TACE）は適切な症例選択の下に標準治療として確立している。一方，化学療法はこれまで多くのレジメンが臨床試験として試みられてきたが，生存期間の延長が確認された標準治療もその位置付けも確立していない。最近，癌の進展，増殖にかかわるさまざまな分子生物学的特徴が明らかになり，それらを標的とした分子標的薬の開発が急速に進歩してきた。肝細胞癌においても従来の化学療法に代わって多くの分子標的薬が試みられてきている。本稿では肝細胞癌の化学療法と分子標的薬の臨床試験について概説する。

肝細胞癌に対する化学療法

肝細胞癌に対する化学療法は，肝動脈から注入する経動脈性化学療法（動注化学療法）と経静脈あるいは経口による全身化学療法に分けられる。肝細胞癌に対する化学療法は，肝切除，局所壊死療法，肝動脈塞栓（化学塞栓）療法の局所治療が無効あるいは適応困難な例（高度門脈腫瘍栓など）および遠隔転移例が適応となる。また肝細胞癌では肝硬変など慢性肝障害を背景にもつ例が多いことから，肝障害を助長するリスクも大きく，肝障害度C（Child-Pugh C）の肝機能不良例では化学療法は禁忌である。

わが国では肝動脈からの動注化学療法が盛んに行われている。現在，動注化学療法剤としてepirubicin，mitomycin C，5-FUが主に用いられてきたが，2004年7月，cisplatin（アイエーコール®）の保険適応が承認された。最近では5-FU＋cisplatinや5-FU＋in-

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表1　切除不能肝細胞癌における doxorubicin と無治療との無作為化比較試験[5]

	doxorubicin	best supportive care	
N	60	46	
response	2 (3.3%)	-	
median OS	10.6 weeks	7.5 weeks	$p=0.036$
fetal complication	15 (25%)		
cause of death			
tumor progression with cachexia	60.0%	76.1%	
side effects of therapy	25.0%	0	
GI bleeding	6.6%	8.7%	
rupture of tumor	3.3%	6.5%	
hypoglycemia	5.0%	4.3%	
subarachnoid hemorrhage	0	2.3%	
suicide	0	2.3%	

表2　切除不能肝細胞癌における doxorubicin と PIAF 併用療法との無作為化第Ⅲ相試験[12]

	doxorubicin	PIAF	p-value
N	94	94	
response	10.5%	20.9%	0.058
median overall survival	6.83 months	8.67 months	0.83
treatment-related mortality	3%	9%	0.194
major toxicity*			
neutropenia	63%	82%	0.003
thrombocytopenia	24%	57%	<0.001
vomiting	4%	12%	0.058
hypokalemia	0%	7%	0.007
hyponatremia	1%	6%	0.054

*Grade 3 or above

terferon（IFN）で高い奏効率が報告されているが，いずれも前向きな臨床試験による検証は行われていない[10,11]。

全身化学療法では，これまで肝細胞癌における無作為化比較試験として doxorubicin（DXR），tamoxifen，interferon などいくつか行われてきた[5-9]。DXR では無治療群に比べ有意に生存期間の延長が得られたが，25%の症例で致命的な合併症が認められている（表1）[5]。tamoxifen や IFN-α では生存期間の改善は認められていない[7-9]。最近では多剤併用療法が試みられ，5-FU/mitoxantrone/cisplatin（FMP），cisplatin/doxorubicin/5-FU/IFN-α（PIAF）などで25%を超える高い奏効率が報告された[10,11]。これらのなかで DXR を control arm とした PIAF regimen の第Ⅲ相試験が行われたが，有意な生存期間の改善は示せず（表2）[12]，既存の抗癌剤による化学療法は悲観的に考えられていた。

上皮成長因子（epidermal growth factor）

細胞の増殖は，epidermal growth factor（EGF），transforming growth factor（TGF）-α などさまざまな成長因子が receptor と結合して生じるシグナル伝達経路を介して制御されている。肝細胞癌でもこれらの receptor の高発現が報告されており，EGF receptor（EGFR）を阻害することにより，癌の増殖，転移，浸潤など進行を止め，アポトーシスを誘導する効果が期待されている[13]。

ヒト EGFR は4種類（human epidermal growth factor receptor：HER1～4）に分けられ，EGFR/

表3 肝細胞癌に対する上皮成長因子受容体阻害薬の主な第Ⅰ，Ⅱ相臨床試験

	class of anti-EGF agent	targets	study design	n	response rate	disease control rate	median PFS	median OS	author	year
erlotinib	tyrosine-kinase inhibitor	HER1/EGFR	Phase Ⅱ	38	9％	59%	3.2mo	13.0mo	Philip	2005[14]
erlotinib			Phase Ⅱ	40	0	43%	3.1mo	6.3mo	Thomas	2007[15]
lapatinib		HER1/EGFR +HER2	Phase Ⅱ	40	5％	38%	2.3mo	6.2mo	Ramanathan	2006[16]
cetuximab	antibody	HER1/EGFR	Phase Ⅱ	32	0	44%	8wk	-	Gruenwald	2007[17]
cetuximab			Phase Ⅱ	30	0	17%	1.4mo	9.6mo	Zhu	2007[18]
GEMOX/ cetuximab			Phase Ⅱ	45	24%	65%	4.5mo	9.2mo	Louafi	2007[19]

GEMOX；gemcitabine + oxaliplatin, response；complete response + partial response, disease control；complete response + partial response + stable disease, PFS；progression-free survival, OS；overall survival

HER1とHER2阻害薬が臨床応用されている。receptor の細胞外のドメインを阻害する抗体薬と細胞内のドメインである tyrosine kinase を直接阻害する阻害薬が開発されている（表3）。これらの薬剤（いわゆる分子標的薬）を用いた臨床試験が多く行われ，肝細胞癌においても EGFR/HER1の高発現は52～71％と高率に認められている。

erlotinib は tyrosine kinase 阻害薬であり，わが国でも肺癌ですでに保険適応の承認が得られている。肝細胞癌での第Ⅱ相試験では奏効率0～9％，無増悪生存期間（PFS）中央値3カ月程度と単独での開発は難しい印象がある[14,15]。cetuximab は IgG1サブクラスのヒト・マウスキメラ化モノクローナル抗体であり，EGFR と結合してその働きを阻害する。これまで単独あるいは gemcitabine＋oxaliplatin（GEMOX）との併用が試みられているが，単独では奏効率0％，PFS 中央値2カ月未満と期待された効果は得られていない[18,19]。今後，これらの EGFR 阻害薬は従来の抗癌剤との併用や後述する血管新生阻害薬との併用などの試みが期待されている。

血管内皮増殖因子（vascular endothelial growth factor）

肝細胞癌は血管新生の豊富な腫瘍の代表であり，血管内皮増殖因子（vascular endothelial growth factor；VEGF）との関係が強いと考えられている。Poon らは肝細胞癌の進行や予後に対する VEGF の関与を報告している[20]。肝細胞癌患者と健常人で血清 VEGF 値を比較すると，肝細胞癌患者245pg/ml，健常者180pg/ml と患者群で有意に上昇していた（p＝0.042）。また肝細胞癌の進行度と血清 VEGF 値を比較すると，サイズ，Edmondson grade，血管浸潤の有無，肝内サテライト病変の有無，TNM 分類では進行した群で血清 VEGF 値が高値であった。VEGF 値と予後との関連では，高 VEGF 群で予後不良であった（p＝0.012）。VEGF の阻害は腫瘍血管の減少，血管の蛇行・ねじれの改善から間質圧の減弱と血管の正常化をもたらすとされる。その結果，癌のアポトーシスを促すことに加え，抗癌剤のデリバリーを高める。このように VEGF は肝細胞癌の進行度や予後，化学療法の効果増強などと大きく関連しており，VEGF 阻害薬は肝細胞癌の治療適応に期待されている。

VEGF 阻害薬として，VEGF を中和する抗体薬 bevacizumab と VEGF receptor（VEGFR）を中心とした複数の標的を阻害するマルチキナーゼ阻害薬，VEGFR をより選択に阻害する VEGFR 阻害薬などが開発されている（表4）。bevacizumab は VEGF と特異的に結合する遺伝子組換え型ヒト化モノクローナル抗体であり，単独，抗癌剤との併用，erlotinib との併用が試みられている[21-23]。とくに bevacizumab と erlotinib 併用療法では奏効率21％，PFS 中央値9.0カ月，全生存期間（OS）中央値19.0カ月と良好な成績が得られており，今後，第Ⅲ相試験の実施が注目される[23]。

分子標的治療としては，癌細胞自体の増殖因子である EGF シグナル伝達路と血管内皮増殖の両方の阻害が理想的である。その観点から複数の分子標的を阻害するマルチキナーゼ阻害薬が開発されてきた。sorafenib は EGFR の下流である RAF [illegible]