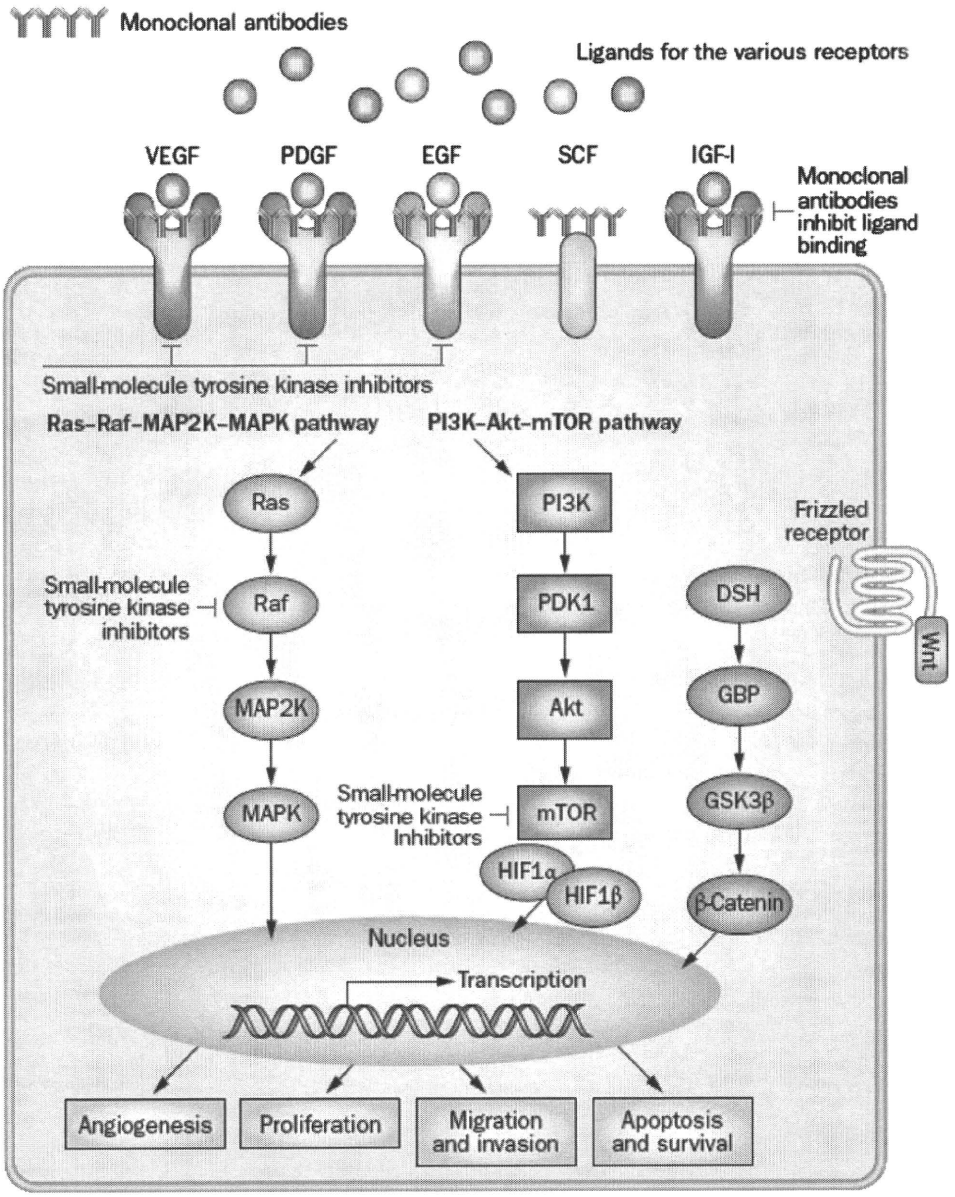

Fig. 1 Signaling pathways and the site of action of molecular-targeted agents (modified from Villanueva A et al. [3] and Llovet and Bruix [5])

are rare, and no *k-ras* or *b-raf* mutations have been detected [16]. On the other hand, wild-type Raf-1 was reported to be hyperactivated in many cancers, including HCC [17–19]. Sorafenib inhibits Raf, and has multiple characteristics in that it has strong inhibitory activity against Raf-1 (C-Raf) kinase, B-Raf (wild-type B-Raf and mutant V600E B-Raf) serine/threonine kinase, the pro-angiogenic receptor tyrosine kinases VEGFR, PDGFR, and FGFR1, and tyrosine kinases such as c-kit, Flt-3, and RET, which are involved in tumor progression and overall prognosis [20].

MEK

The MEK family consists of MEK1 and MEK2 proteins, which specifically phosphorylate tyrosine and threonine residues, and phosphorylates downstream Erk1 and Erk2 [21].

In an immunohistochemical study, MEK1/2 overexpression, ERK1/2 overexpression, and ERK1/2 phosphorylation were observed in 100% (46/46), 91% (42/46), and 69% (32/46) of HCCs, respectively, and the in-vitro treatment of HepG2 and Hep3B cells with MEK1/2 inhibitors inhibited cell growth and upregulated apoptosis [22].

The MEK inhibitors CI-1040, PD0325901, AZD6244, and RDEA119/BAY869766 have been tested in several cancers including solid tumors such as HCC. A phase II study of AZD6244 (selumetinib, ARRY-142866) and a phase I/II study of RDEA119/BAY869766 in combination with sorafenib are being conducted.

Table 1 Molecular-targeted agents being tested in HCC

Agent	Antiangiogenic targets					Antiproliferative targets				Antiepigenetic targets				Developmental status	Company
	VEGF	FGF	VEGFR	PDGFR	FGFR	EGFR	Raf	MEK	mTOR	RAR	RXR	HDAC	Heparanase		
Sorafenib[a] (Nexavar)			●	●			●							Approved	Bayer
Sunitinib[a] (Sutent)			●	●										Phase III stopped	Pfizer
NIK-333 (Acyclic Retinoid)										●	●			Phase II/III complete	Kowa
Brivanib			●		●									Phase III ongoing	Bristol-Myers Squibb
TSU-68			●	●	●									Phase II complete	Taiho
TAC-101										●				Phase II stopped	Taiho
Erlotinib (Tarceva)						●								Phase II complete	Roche
Bevacizumab (Avastin)	●													Phase II ongoing	GenentechA
AZD2171 (Cediranib)			●											Phase II recruiting	AstraZeneca
Gefitinib (Iressa)						●								Phase II complete	AstraZeneca
Lapatinib						●								Phase II ongoing	GlaxoSmithKline
Thalidomide	●													Phase II ongoing	TTY BioPharm
Linifanib			●	●										Phase III initiated	Abott
AZD6244								●						Phase II ongoing	AstraZeneca
PI-88	●	●											●	Phase II complete	Progen
Cecuximab						●								Phase II complete	Merck
RAD001									●					Phase III initiated	Novartis
PXD101 (Belinostat)												●		Phase I/II ongoing	Curagen

Sources: Trial Trove, ClinicalTrials.Gov (NCI), Evaluate Pharma, IMS Knowledge Link, Esplcom, IDdB3, BioPharm Insight, MedTrack

[a] Sorafenib and sunitinib also have antiproliferative affects through multi-tyrosine kinase inhibition

PI3K/Akt/mTOR pathway

The PI3K/Akt/mTOR pathway also plays an important role in cell growth, survival regulation, metabolism, and anti-apoptosis. The membrane lipid phosphatidylinositol 4,5-bisphosphate (PIP_2) is phosphorylated by phosphatidylinositol 3-kinase (PI3K) into phosphatidylinositol 3,4,5-triphosphate (PIP_3), which binds to and activates the serine/threonine kinase Akt. The tumor-suppressor gene product PTEN (phosphatase and tensin homolog deleted on chromosome) is antagonistic to PI3K activity. PTEN is a lipid phosphatase that dephosphorylates inositol phosphates such as PIP_3. The inactivation of PTEN through gene deletion increases PIP_3 levels, and activates Akt, which inhibits apoptosis, leading to the development of tumors. The serine/threonine kinase mTOR is an important mediator in the PI3K/Akt pathway that binds intracellularly to a protein called raptor or rictor, and exists as two different complexes, complex 1 and 2 (mTORC1 and mTORC2). mTORC2 (mTOR-rictor) activates Akt whereas mTORC1 (mTOR-raptor) is activated downstream of Akt; thus, both molecules regulate protein synthesis [23].

A study of 528 HCC samples showed that expression of pAkt, PTEN, p27, and S6 ribosomal protein (pS6) was a poor prognostic factor for survival [24]. A tissue microarray analysis of HCC samples revealed that the loss of PTEN and overexpression of pAkt and p-mTOR were correlated with tumor grade, intrahepatic metastasis, vascular invasion, TNM stage, Ki-67 labeling index, and matrix metalloproteinase (MMP)-2 and (MMP)-9 upregulation, Meanwhile, PTEN mRNA expression in the cancerous tissue was downregulated, compared with that in the non-cancerous tissue. The levels of PTEN, MMP-2, and MMP-9 mRNA expression were correlated with tumor stage and metastasis, and the levels of PTEN and MMP-9 mRNA expression were inversely correlated [25]. In an extensive analysis of 314 HCC samples in terms of mutation analysis, DNA copy number changes, mRNA levels, and immunostaining, Villanueva et al. found that activation of the IGF pathway, upregulation of EGF, dysregulation of PTEN, and aberrant mTOR signaling were present in half of the samples, and that inhibiting mTOR activity with everolimus was effective in improved survival and suppression of recurrence [26].

The PI3K inhibitor RG7321 and the Akt inhibitor perifosine target the PI3K/Akt/mTOR pathway and are in early stages of clinical development, whereas the mTOR inhibitors everolimus (RAD001), sirolimus (Rapamune), and temsirolimus (CCI-779) are at more advanced stages of development. Everolimus is used to treat sorafenib-intolerant patients or for patients showing disease progression after sorafenib administration. A phase III study to compare everolimus and a placebo (EVOLVE-1: Advanced Hepatocellular Carcinoma after Disease Progression or Intolerance to Sorafenib *Ever*O*l*imus for *LiV*er cancer *E*valuation) and a phase I/randomized phase II study (sorafenib + everolimus vs. sorafenib alone) to test the efficacy and tolerance of sorafenib in combination with everolimus are underway. Because mTOR inhibitors have cytostatic and antiangiogenic effects, they are expected to be effective in combination with other angiogenesis inhibitors such as bevacizumab, and may be appropriate for administration after transarterial chemoembolization (TACE). Furthermore, because the mTOR pathway is stimulated by factors such as EGFR, PDGFR, and TGFα, and is closely related to other signaling pathways including the Ras/Raf/MEK/ERK pathway, they are likely to show promising efficacy when used in combination with other growth factor inhibitors [27].

VEGF/VEGFR, PDGFR, FGFR

Angiogenesis is an important event not only for HCC but also for cancer growth and metastasis, and occurs because of complex alterations involving promoting factors such as VEGF, angiopoietin, and FGF, inhibitory factors including thrombospondin (TSP) and angiostatin and the surrounding tissue. The VEGF family consists of VEGF-A, B, C, D, and E, and placental growth factor (PIGF). The VEGFR family comprises VEGFR-1 (flt-1), VEGFR-2 (flk-1/KDR), and VEGFR-3 (flt-4). VEGF-A binds to VEGFR-1 and VEGFR-2 and is involved in angiogenesis and the maintenance of mature blood vessels, whereas VEGF-C and VEGF-D mainly bind to VEGFR-3, are involved in lymphangiogenesis [28, 29]. VEGF isoforms such as $VFGF_{121}$ and $VEGF_{165}$ have been identified, and isoform subtypes also exist, for example EGF_{166}b. Thus, it is clear that these growth factors do not exhibit angiogenesis-promoting effects alone, and they have attracted attention as new therapeutic targets [30].

HCC typically exhibits active angiogenesis. During the progression from early to well, and to moderately differentiated HCC, angiogenesis increases and cancer cells acquire the ability to invade vessels and metastasize. Scientific and clinical studies have revealed that, during the progression from hepatitis to cirrhosis, angiogenesis and disruption of the vascular architecture are linked to the progression of HCC, and contribute to increased hepatic vascular resistance and portal hypertension, and decreased hepatocyte perfusion [31]. In addition, a meta-analysis has demonstrated that VEGF expression is a prognostic factor in HCC [32].

Phase II studies have been started to test the usefulness of bevacizumab (Avastin®), which directly targets VEGF, in TACE-treated HCC, and the use of bevacizumab in

combination with erlotinib (Tarceva®), an EGFR tyrosine kinase inhibitor.

Sunitinib (Sutent®) is a multi-kinase inhibitor that inhibits tyrosine kinases such as VEGFR-1, 2, 3, PDGFR-α, β, and c-Kit. A phase II study of sunitinib in 37 advanced HCC patients showed that the median progression-free survival (PFS) and median overall survival (OS) were 3.7 and 8 months, respectively. In that study, adverse events included grade 3/4 thrombocytopenia in 37.8% of patients, neutropenia in 24.3%, asthenia in 13.5%, and hand–foot syndrome in 10.8% [33]. Because sunitinib has a lower IC_{50} for each target than sorafenib, it is expected to have greater antitumor activity. However, this factor may be responsible for the higher incidence of adverse events with sunitinib. The main evaluation item in the above phase II trial was the response rate, which did not reach the expected value, leading to the conclusion that it was a negative study [34]. In that study sunitinib was administered at 50 mg/day for 4 weeks followed by 2 weeks of rest per cycle [33], whereas Zhu et al. [34] used a dosing schedule of 37.5 mg/day for 4 weeks followed by 2 weeks of rest per cycle, and reported that the median PFS and OS were 3.9 and 9.8 months, respectively. An ongoing global cooperative phase III controlled clinical trial to compare sorafenib and sunitinib head-to-head and to seek approval for first-line indications for advanced HCC adopted a sunitinib dosing schedule of 37.5 mg/day. However, in a "Reflection and Reaction" regarding these trial results, Forner et al. cast doubt on whether the drugs at this dose could maintain tolerance and ensure efficacy [35]. Because recruitment is progressing well, the results are expected to be available soon.

Brivanib is a kinase inhibitor that selectively inhibits VEGFR-1, 2, and 3, and FGFR-1, 2, and 3. As for sunitinib, an international global phase III clinical trial to compare brivanib and sorafenib head-to-head and to seek approval for first-line therapy for advanced HCC has already been started, and the results are eagerly awaited. Japanese centers are participating in this clinical trial. Because brivanib targets FGF and VEGF, and is associated with relatively mild adverse effects, a second-line study of brivanib in sorafenib-ineffective and sorafenib-intolerant patients and a trial to evaluate the use of brivanib in combination with TACE are underway. Depending on the results of these trials, indications for use in HCC may be obtained; therefore, positive results are eagerly anticipated. The results have been reported for a phase II study of brivanib in 55 patients (cohort A) who had not received systemic therapy for curatively unresectable HCC and 46 patients (cohort B) previously treated with angiogenesis inhibitors such as sorafenib or thalidomide [36]. The median TTP and OS were 2.8 and 10 months, respectively, in cohort A versus 1.4 and 9.8 months, respectively, in cohort B. Adverse events included fatigue (51.5%), diarrhea (41.6%), hypertension (42.6%), anorexia (41.6%), and nausea/vomiting (40.6/30.7%). Thus, these results demonstrated the efficacy of brivanib as a second-line treatment. The results of three phase III clinical trials, BRISK-PS (sorafenib failure or sorafenib-intolerant patients; brivanib + best supportive care (BSC) vs. placebo + BSC), BRISK-FL (advanced HCC; brivanib vs. sorafenib), and BRISK-TA (patients with unresectable HCC, brivanib vs. placebo as post-TACE adjuvant therapy) are awaited. Japanese centers participated in all three trials.

In a Japanese phase I/II trial of TSU-68, an oral molecular inhibitor of VEGFR, PDGFR, and FGFR, to test its safety and efficacy in 35 HCC patients, the response rate was 5.6% (CR, PR, SD, PD, and NE in 1, 2, 15, 16, and 1 patients, respectively), and the disease control rate was 51.4% [37].

In addition, several phase I/II trials are being conducted to assess kinase inhibitors such as linifanib (ABT-869) and cediranib (AZD2171), which inhibit VEGFR, PDGFR, CSF-1R (cFms), Kit, and Flt3. Furthermore, axitinib, which is currently being tested in renal cell carcinoma, has also attracted attention as a promising agent for treatment of HCC because of its efficacy and mild side effects.

EGF/EGFR

EGFR is a member of the human epidermal growth factor receptor (HER) family that includes EGFR (erbB1), HER2/neu (erbB3), and HER4 (erb4). All members of this family, except HER3, have an intracellular tyrosine kinase domain, and the binding of a ligand to its extracellular domain triggers signal transduction through the above-described MAPK and PI3K/Akt/mTOR pathways. Thus, these receptors are involved in cell growth, differentiation, survival and adhesion [38]. EGFR overexpression has been reported in many cancers, and in HCC. For example, Buckley et al. reported that EGFR, detected by immunohistochemical analysis, was overexpressed in 50 (66%) of 76 HCCs, and that fluorescence in-situ hybridization (FISH) showed extra EGFR gene copies in 17 (45%) of 38 HCCs [39].

EGFR-targeting drugs, which include anti-EGFR antibodies, such as cetuximab and panitumumab, and small-molecule inhibitors of EGFR tyrosine kinases such as gefitinib, etc., have been used widely for treatment of several cancers other than HCC. Unfortunately, except for phase II trial data, there are few clinical data on the efficacy of these drugs for the treatment of HCC.

Similar to gefitinib (Iressa®), erlotinib (Tarceva®) is an oral EGFR tyrosine kinase inhibitor. Philip et al. and Thomas et al. have reported the results of phase II studies of erlotinib in HCC [40, 41]; the median OSs in their

studies were 13 and 10.7 months, respectively. A phase III clinical study (SEARCH study: *S*orafenib and *E*rlotinib, a *R*andomized T*r*ial Protoc*o*l for the Treatment of Patients with *H*epatocellular Carcinoma) of sorafenib in combination with erlotinib versus sorafenib plus placebo is ongoing. Because erlotinib is associated with a high incidence of skin rash, dry skin, and gastrointestinal toxicity, for example diarrhea, the results of the SEARCH study should be evaluated to assess whether this combination therapy can be used in clinical settings. Thomas et al. conducted a phase II clinical study of erlotinib in combination with bevacizumab in 40 advanced HCC patients, and reported promising results; the median PFS and OS were 9 and 15.7 months, respectively. However, they noted frequent treatment-related grade 3/4 toxicities, including fatigue (20%), hypertension (15%), gastrointestinal bleeding (12.5%), wound infection (5%), diarrhea (10%), elevated transaminase levels (10%), and thrombocytopenia (10%) [42], which necessitates further evaluation of drug tolerance. Although a clinical study of erlotinib in combination with bevacizumab (OPTIMOX-3 study) was also conducted in colorectal cancer patients, no tolerance was observed, which led to a change in the protocol [43, 44].

After the introduction of a number of molecular-targeted drugs, strategies for the inhibition of similar or different signaling pathways (vertical or horizontal inhibition) with several drugs have been proposed. However, the combined use of molecular-targeted agents has remained largely unsuccessful, including panitumumab in combination with bevacizumab for treatment of colorectal cancer [45]. Similarly, results for sorafenib in combination with bevacizumab (vertical inhibition) have been reported [46]. Although some therapeutic response was obtained, the combination therapy resulted in greater toxicity [46], suggesting the need for detailed evaluation of the dosing regimen.

Lapatinib (Tykerb®) is a dual inhibitor of EGFR and HER-2/neu, and inhibits tumor growth by downregulating MAPK, AKT, and p70S6 kinase [47]. In Japan, lapatinib is indicated for treatment of breast cancer. In a phase II clinical trial of lapatinib in 26 patients with unresectable advanced HCC, the median PFS and OS were 1.9 and 12.6 months, respectively, and adverse events included diarrhea (73%), nausea (54%), and skin rash (42%) [48].

Cetuximab (Erbitux®) is a human/mouse chimeric monoclonal antibody consisting of the variable region of a mouse anti-human EGFR monoclonal antibody and the human IgG1 constant region. Cetuximab inhibits the binding of endogenous EGFR ligands, for example EGF and TGFα, to EGFR. In a phase II clinical trial of cetuximab in 30 patients with unresectable or metastatic HCC, the median PFS and OS were 1.4 and 9.6 months, respectively, and treatment-related toxicities included grade 3 hypomagnesemia (3.3%) and grade 1/2 acne-like rash (83.3%), which was observed for the duration of anti-EGFR therapy in that study [49].

The EGFR is a very interesting therapeutic target. As described above, use of erlotinib in combination with sorafenib is still in the research stage. However, on the basis of results from phase II studies, the efficacy of cetuximab or lapatinib as monotherapy seems to be limited, and the results of further studies evaluating their efficacy in sorafenib-refractory or intolerant patients are awaited with interest.

HGF/c-Met pathway

Because the hepatocyte growth factor (HGF)/Met pathway is involved in tumor growth, invasion, and angiogenesis in a wide range of neoplasms, HGF and Met have recently attracted attention as therapeutic targets. HGF is a heterodimer consisting of α and β chains bound together by a disulfate bond. The α chain contains four kringle domains, and the β chain contains a serine protease-like domain. Met is a receptor tyrosine kinase for the HGF ligand, and contains a semaphorin-like domain. HGF or Met overexpression and Met gene mutations and duplications have been reported in various cancers, and abnormalities due to HGF/Met pathway activation have also been noted [50]. These abnormalities activate the downstream signaling cascade, leading to epithelial-mesenchymal transition and increased proliferative, migratory, invasive and metastatic potentials of cancer cells [50].

HGF/c-MET-targeted drugs, including kinase inhibitors, HGF inhibitors, and decoy c-Met receptor molecules, are being developed. Of particular interest is ARQ-197, a c-Met receptor tyrosine kinase inhibitor which is a non-ATP-competitive molecule that binds near the ATP-binding site. A randomized phase II study of ARQ-197 versus placebo is ongoing in patients with unresectable HCC after systemic therapy failure.

IGF/IGFR

The IGF/IGFR system is involved in cell growth and the chemotherapeutic response. The ligands IGF-I and II bind to their receptors IGF-1R and IGF-2R, and are involved in DNA synthesis and cell growth. Abnormalities in IGF and IGF-1R or their overexpression have been reported in various cancers, including HCC. Their associations with disease stage, metastasis, and survival [51] and the functions of IGF and IGFR in HCC [52] have been reported.

IGF-targeting drugs are currently being developed, and mainly include anti-IGF-1R antibodies, for example BIIB022, AVE1642, and cixutumumab (IMC-A12). A phase II study of cixutumumab, a phase Ib/II study of

Table 2 Results of the Asia–Pacific and SHARP studies

End point	Asia–Pacific		SHARP	
	Hazard ratio (95% CI)	*P* value	Hazard ratio (95% CI)	*P* value
OS	0.68 (0.50–0.93)	0.014	0.69 (0.55–0.87)	<0.001
TTSP	0.90 (0.67–1.22)	0.498	1.08 (0.88–1.31)	0.768
TTP	0.57 (0.42–0.79)	<0.001	0.58 (0.45–0.74)	<0.001
PFS	0.62 (0.46–0.82)	<0.001	0.65 (0.52–0.79)	<0.001

sorafenib versus sorafenib plus BIIB022, and phase I/II studies of AVE1642 as monotherapy or in combination with sorafenib or erlotinib are ongoing.

Sorafenib: trial results and clinical experience

Clinical results for sorafenib in HCC

As described above, sorafenib is a multi-kinase inhibitor of tumor growth and angiogenesis, and has a strong inhibitory effect on C-Raf and B-Raf serine/threonine kinases (comprising the Raf/MEK/ERK pathway), VEGFR and PDGFR tyrosine kinases, and FLT-3 and c-kit [20]. To date, sorafenib is the only molecular-targeted agent approved for treatment of HCC, on the basis of the results of two large-scale clinical trials, namely the SHARP (Sorafenib HCC assessment Randomized Protocol) study [53] and the Asia–Pacific study [54]. The median OSs for the sorafenib group in the SHARP and Asia–Pacific studies were 10.7 months (vs. 7.9 months for the placebo group, $P < 0.001$; HR: 0.69) and 6.5 months (vs. 4.2 months for the placebo group, $P = 0.014$; HR: 0.68), respectively, indicating that sorafenib prolongs survival by approximately 50% (Table 2). These data should compel HCC specialists to challenge their preconception that systemic anticancer drug therapy is not effective for HCC.

Current status regarding the use of sorafenib in Japan

Sorafenib was approved in Japan in May, 2009. A survey has confirmed that, at the time of writing (March, 2010), over 3,700 patients have been prescribed sorafenib. Across several centers, 15 Japanese patients have achieved CR, which was not observed in the SHARP or Asia–Pacific trials. This suggests that some Japanese patients may be very sensitive to sorafenib [55]. The reason for this, and predictive biomarkers, are now actively under investigation.

On the other hand, it has been reported that hand–foot syndrome occurs early after sorafenib administration [56] more often than was noted in the SHARP and Asia–Pacific studies, and the drug is often discontinued because of the adverse effects in many patients [56]. As demonstrated in the SHARP and Asia–Pacific studies, sorafenib is only used to achieve stable disease; it is, therefore, important to improve drug efficacy by extending the period of administration for as long as possible. Therefore, it is no exaggeration to say that, in the case of sorafenib, the "successful management of side effects" is equal to "successful treatment." According to "post-TACE phase III clinical study [56]" performed in Japan and Korea, it is strongly speculated that physicians who are unaccustomed to prescribing molecular-targeted agents and who fail to see marked efficacy, as induced by conventional chemotherapeutic agents, often do not understand the properties of this drug, and they (and the patients) do not fully comprehend therapeutic efficacy. Moreover, they feel too anxious about side effects that have not been encountered before. These circumstances may result in treatment discontinuation in many patients. Clearly, greater awareness among physicians for therapeutic efficacy and approaches to manage adverse effects is needed to improve treatment outcomes.

Experience of sorafenib use at our institute

Since the approval of sorafenib on May 20, 2009, we have treated 90 patients with sorafenib, and few have discontinued therapy because of adverse effects or patient refusal to continue. Of these 90 patients, two achieved CR [55]. These two CR patients, in whom pulmonary and adrenal metastases and intrahepatic lesions all disappeared, survived free of recurrence for more than 2 years and 1 year, respectively, at the time of writing (March, 2010), i.e., they are still alive at present. In other patients who apparently achieved SD, the tumor marker levels reached a plateau after sorafenib administration, when their levels were rising rapidly before sorafenib administration. Even if hepatic lesions do not show a clear tendency to undergo necrosis or regression on CT images, three tumor markers (AFP, PIVKA-II, and AFP-L3) are widely considered to serve as surrogate markers. In fact, there are very few data on serum tumor markers, except for AFP, outside Japan. Nevertheless, Japanese researchers have demonstrated the value of changes in these markers and the antitumor efficacy of sorafenib [55].

Interestingly, it has previously been demonstrated that the levels of PIVKA-II or DCP tend to be increased by

inducing hypoxia [57]. Therefore, PIVKA-II or DCP may be a good predictive marker for evaluating the hypoxic response to antiangiogenic therapy for HCC.

Only 17 of the 90 patients showed PD on computed tomography (CT) images although follow-up period is still short (less than 10 months). However, because the speed with which the patient develops progressive disease may slow down due to tumor growth inhibition, it is very difficult to determine when to discontinue treatment because of tumor refraction. Important issues for future studies include:

1 identification of biomarkers that can be used to predict therapeutic responses, including CR or PR, in patient groups;
2 evaluation of the role of tumor markers in the determination of therapeutic responses;
3 establishing response evaluation criteria that can determine the therapeutic responses to molecular-targeted agents; and
4 development of effective second-line therapies after sorafenib failure (Figs. 2, 3).

In the treatment algorithm (Figs. 2, 3) approved by the Consensus Meeting of the 2009 Annual Meeting of the Japan Society of Hepatology (Congress chair: Professor Masatoshi Kudo), sorafenib is indicated for treatment of patients with Child-Pugh A HCC with extrahepatic metastasis, vascular invasion, or refractoriness to TACE or arterial infusion chemotherapy.

In addition to the pharmaceutical-sponsored clinical trials of sunitinib and brivanib as first and second-line therapy in sorafenib-refractory patients, investigator-initiated trials (IIT) of sorafenib in combination with hepatic arterial infusion chemotherapy (SILIUS trial), pharmaceutical and IIT trials of sorafenib in combination with TACE (SPACE, TACICS and BRISK-TA trials), and a trial to test the inhibitory effect of sorafenib on tumor recurrence after curative treatment (STORM trial) are ongoing, and the results of these trials are eagerly awaited (Figs. 2, 3). The working hypotheses in these studies can be deduced by extrapolating the MST and hazard ratios in overall survival (OS) calculated in a subanalysis of the SHARP study (Table 3). The results obtained suggest that starting treatment with molecular-targeted drugs at an earlier tumor stage in combination with standard treatment options such as resection, ablation, TACE, or hepatic arterial infusion chemotherapy can improve the prognosis of HCC. Thus, sorafenib has the potential to induce a change of emphasis in the treatment of HCC. For example, in a subanalysis of the SHARP trial, the hazard ratios for OS and MST ratio in intermediate stage HCC without vascular invasion or extrahepatic spread were 0.52 and 1.50, respectively (Table 4). This suggests that survival of early stage HCC and intermediate stage HCC may be prolonged from 5 years to 7.5–10 years by using sorafenib in an adjuvant setting after curative treatment, and from 3 years to 4.5–6 years by using sorafenib in combination with TACE (Fig. 4).

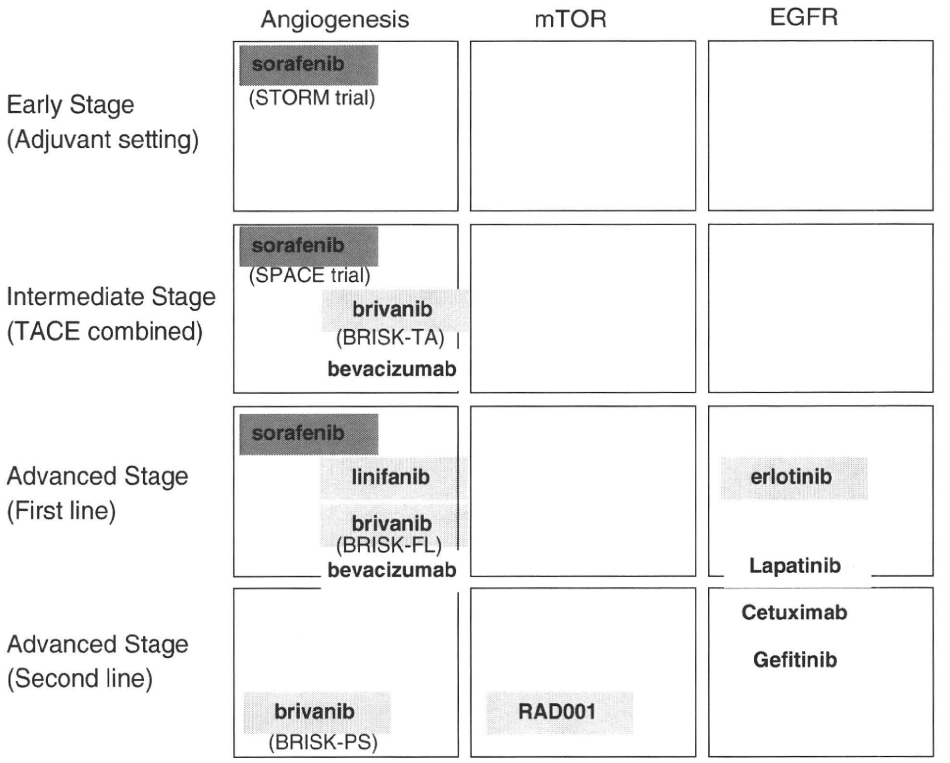

Fig. 2 Molecular targeted agents: ongoing trials in each stage of HCC

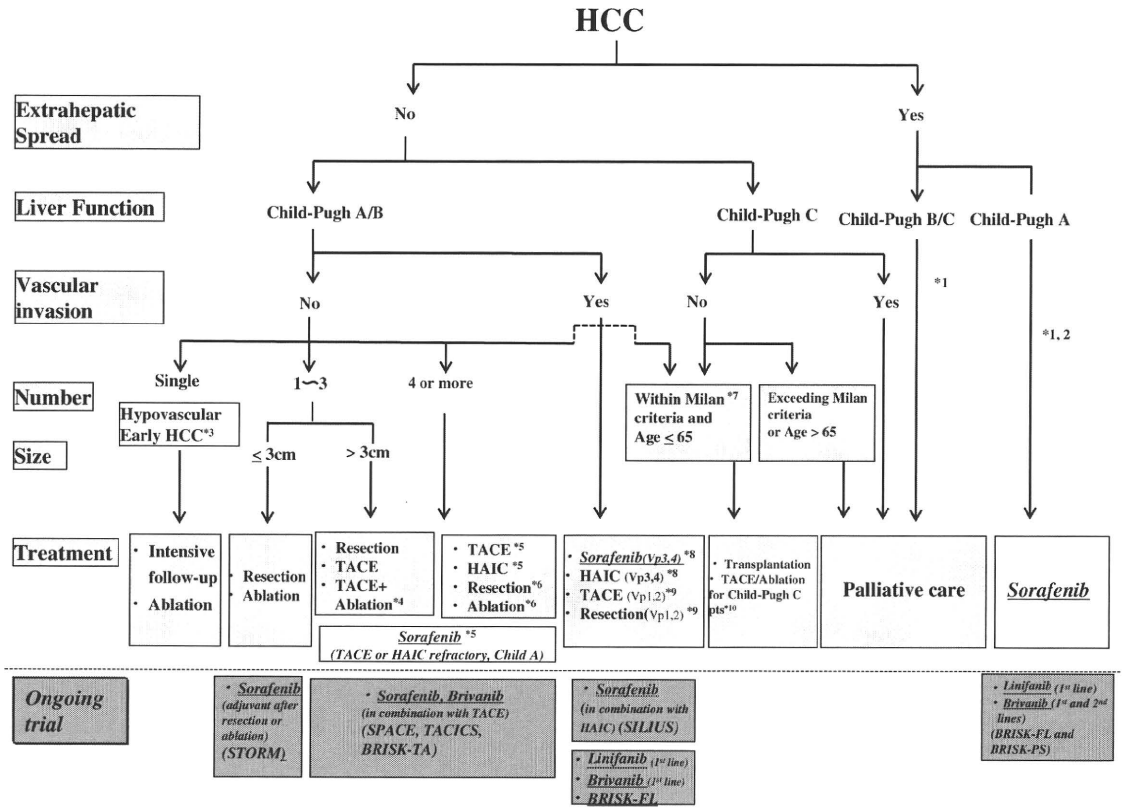

Fig. 3 Consensus-based treatment algorithm for HCC proposed by the Japan Society of Hepatology (JSH) revised in 2010. *1 Treatment should be performed as if extrahepatic spread is negative, when extrahepatic spread is not considered as a prognostic factor in Child-Pugh class A/B patients, *2 sorafenib is the first choice of treatment in this setting as a standard of care, *3 intensive follow-up observation is recommended for hypovascular nodules by the Japanese Evidence-Based Clinical Practice Guidelines. However, local ablation therapy is frequently performed in the following cases: (1) when the nodule is diagnosed pathologically as early HCC; (2) when the nodules show decreased uptake on Gd-EOB-MRI; or (3) when the nodules show decreased portal flow by CTAP, since these nodules frequently progress to advanced HCC, *4 even for HCC nodules exceeding 3 cm in diameter, transcatheter arterial chemoembolization (TACE) in combination with ablation is frequently performed when resection is not indicated, *5 TACE is the first choice of treatment in this setting. Hepatic arterial infusion chemotherapy (HAIC) using an implanted port is also recommended for TACE-refractory patients. The regimen for this treatment is usually low-dose FP (5FU+CDDP) or intra-arterial 5FU infusion combined with systemic interferon therapy. Sorafenib is also recommended for TACE- or HAIC-refractory patients with Child-Pugh class A liver function, *6 resection is sometimes performed when more than four nodules are detected. Ablation is sometimes performed in combination with TACE, *7 Milan criteria: tumor size ≤3 cm and tumor number ≤3, or solitary tumor ≤5 cm. Even when liver function is good (Child-Pugh A/B), transplantation is sometimes considered for patients with frequently recurring HCC, *8 sorafenib and HAIC are recommended for HCC patients with major portal invasion such as Vp3 (portal invasion in the first portal branch) or Vp4 (portal invasion in the main portal branch), *9 resection and TACE are frequently performed when portal invasion is minor, such as Vp1 (portal invasion in the third or more peripheral portal branch) or Vp2 (portal invasion in the second portal branch), *10 local ablation therapy or subsegmental TACE is performed even for Child-Pugh C patients when transplantation is not indicated, when there is no hepatic encephalopathy, no uncontrollable ascites, and a low bilirubin level (<3.0 mg/dl). However, it is regarded as an experimental treatment because there is no evidence of a survival benefit in Child-Pugh C patients. A prospective study is necessary to clarify this issue

Table 3 Subanalysis data of the SHARP study

		Advanced HCC with vascular invasion and extrahepatic spread	Advanced HCC without vascular invasion or extrahepatic spread
Hazard ratio		0.77 (95% CI: 0.60–0.99)	0.52 (95% CI: 0.32–0.85)
Median OS (MST)	Sorafenib	8.9 M (n = 209) (95% CI: 7.6–10.3 M)	14.5 M (n = 90) (95% CI: 14.0 M–N/E)
	Placebo	6.7 M (n = 212) (95% CI: 5.2–8.0 M)	10.2 M (n = 91) (95% CI: 8.6–15.5 M)

M, month

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Table 4 Results of studies of molecular-targeted agents for HCC

Agent	Type	Target	Number of patients	RR (%)	PFS (month)	TTP (month)	OS (month)	Reference
Phase III								
Sorafenib	s.m.	C-Raf, B-Raf,	602 (299[a])	2	–	5.5	10.7	Llovet [5, 53]
		PDGFR, VEGFR	271 (150[a])	3.3	–	2.8	6.5	Cheng [54]
Phase II								
Sorafenib	s.m.	C-Raf, B-Raf, PDGFR, VEGFR	137	2.2	–	5.5	9.2	Abou-Alfa [58]
Sunitinib	s.m.	VEGFR, PDGFR,	37	2.7	3.7	5.3	8	Faivre [33]
		SCFR, FLT3	34	2.9	3.9	4.1	9.8	Zhu [34]
Brivanib	s.m.	VEGFR, FGFR	55	n.r.	–	2.8	10	Raoul [36]
Linifanib	s.m.	VEGFR, PDGFR	44	6.8	–	5.7	9.3	Toh [59]
Bevacizumab	MoAb	VEGF	46	13	6.9	–	12.4	Siegel [60]
Erlotinib	s.m.	EGFR	38	9	–	3.2	13	Philip [40]
			40	0	–	–	10.7	Thomas [41]
Gefitinib	s.m.	EGFR	31	3.2	2.8	–	6.5	O'Dwyer [61]
Lapatinib	s.m.	EGFR	40	5	2.3	–	6.2	Ramanathan [62]
			26	0	1.9	–	12.6	Bekaii-Saab [48]
Cetuximab	MoAb	EGFR	30	0	1.4	–	9.6	Zhu [49]

[a] Sorafenib arm

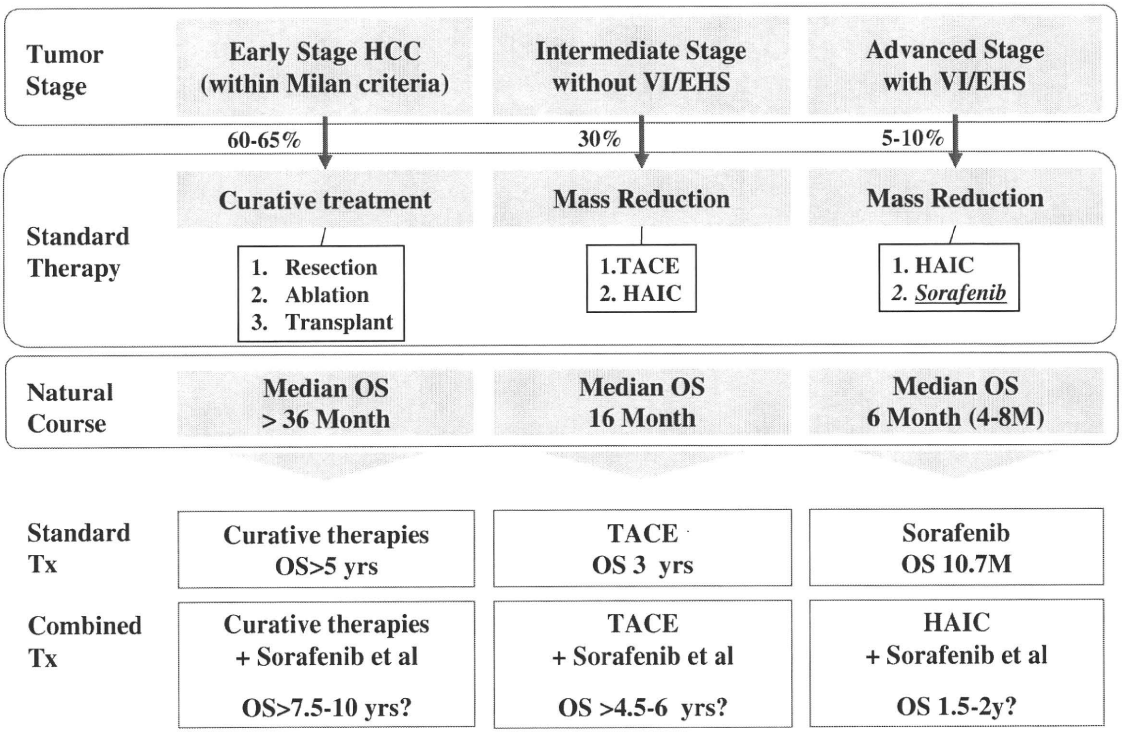

Fig. 4 Outcomes of standard treatment modalities and expected effects of combination therapy with molecular-targeted agents

Summary and future prospects

The results of clinical trials [33, 34, 36, 40, 41, 49, 58–62] of the molecular-targeted agents described above are summarized in Table 4. Angiogenesis-inhibiting drugs, particularly sorafenib, have been evaluated for HCC, and drugs targeting EGFR and mTOR are being developed. The results (numerical values) of phase II clinical trials show no marked differences in the therapeutic efficacy evaluated by time to progression (TTP) or progression-free survival

(RFS). However, results from phase II studies may be subject to patient selection bias and cannot be compared with results from other trials. Thus, when determining the therapeutic efficacy of drugs, we should review the efficacy of the respective drugs, and consider where the theoretical target molecules are present and what combinations of drugs have a theoretical rationale, and thus evaluate options for monotherapy and combination therapy based on the efficacy and safety data obtained from phase III clinical trials.

Molecular-targeted agents that have been introduced into clinical use in recent years are approved for treatment of specific cancer and are then frequently used to treat other cancers. Although not discussed here, studies to identify predictors of efficacy (i.e., biomarkers) for angiogenesis inhibitors and EGFR tyrosine kinase inhibitors, and factors involved in drug resistance, are making steady progress, and the associated therapeutic strategies are undergoing major changes. Therefore, even in the treatment of HCC, it is necessary for HCC specialists to expand their knowledge of and techniques for applying existing treatment modalities (resection, ablation, TACE, arterial infusion chemotherapy) to physically remove, destroy, or necrotize the tumor, and to better understand clinical oncology, particularly the role and mechanisms of action of molecular-targeted agents. We are entering an era in which physicians treating HCC should pay close attention to the development of therapeutic agents not only for HCC but also for other cancers, and be aware of the use of molecular-targeted agents for treating cancers in clinical and basic research settings, and understand approaches to limit or control adverse effects associated with these drugs.

Although sorafenib was recently approved, many issues remain to be addressed, including:

1 how to determine and define refractoriness; and
2 whether to continue TACE or hepatic arterial infusion chemotherapy (a de facto standard in Japan) in patients with TACE-refractory tumors or portal tumor thrombi before starting sorafenib therapy.

For oncology, in particular, the Pharmaceuticals and Medical Devices Agency (PMDA) in Japan has approved several drugs based on results from global clinical trials and on Japanese phase I study data alone. We strongly recommend that, on the basis of the molecular-targeted agents currently under development, clinical studies (including IITs) should be conducted aggressively, and therapeutic strategies should be devised to resolve the limitations of currently used therapeutic approaches and to improve therapeutic outcomes.

The introduction of sorafenib to treat HCC in 2007 in Western countries and in 2009 in Japan was undoubtedly the beginning of a change of emphasis, representing a significant breakthrough for HCC treatment not previously experienced for this unique tumor.

Conflict of interest statement M. Kudo has received honoraria for the lecture from Bayer HealthCare, Pfizer, and Bristol-Meyers.

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INTERNAL MEDICINE

□ CASE REPORT □

Autoimmune Thrombocytopenic Purpura during Pegylated Interferon α Treatment for Chronic Hepatitis C

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Abstract

We describe a 72-year-old woman with chronic hepatitis C and autoimmune thrombocytopenic purpura (AITP) during pegylated interferon (PEG-IFN) α. Immunoglobulin G and antinuclear antibody were 2,113 mg/dL and 1,280 at the start, respectively. A liver biopsy negated autoimmune hepatitis. After a 48-week combination therapy with ribavirin, PEG-IFN α-2a was administered. At the 30th month, the platelet count was decreased to 1.1×10^4/μL. Bone marrow biopsy disclosed normocellular marrow compatible with AITP. The platelet-associated IgG (PAIgG) titer rose to 500 ng/10^7 cells. Corticosteroid therapy was successful, and the platelet count and PAIgG titer reverted to 6.4×10^4/μL and 57.3 ng/10^7 cells, respectively.

Key words: autoimmune thrombocytopenic purpura, interferon, platelet-associated immunoglobulin G, steroid therapy, chronic hepatitis C, immunological disorder

(Inter Med 49: 1119-1122, 2010)
(DOI: 10.2169/internalmedicine.49.3413)

Introduction

A number of autoimmune disorders attributed to interferon treatment such as thyroid disease and diabetes and others have been reported, among which are blood cell-related disorders including red blood cells and platelets. Here, we describe a case of autoimmune thrombocytopenic purpura (AITP) during pegylated interferon (PEG-IFN) α therapy.

Case Report

A 72-year-old woman (149 cm tall, weighing 54 kg) with chronic hepatitis C genotype 1b infection was started on PEG-IFN α-2b (80 μg/week) and ribavirin (600 mg/day) (PEG-IFN/RBV) in July 2006. Laboratory values were as follows: aspartate aminotransferase (AST) 43 (normal, 8-38) IU/L, alanine aminotransferase (ALT) 25 (4-43) IU/L, γ-glutamyl transpeptidase 47 (≤48) IU/L, bilirubin 0.6 (0.2-1.2) mg/dL, hepatitis C virus (HCV)-RNA 2,400 KIU/mL, hemoglobin 12.4 (11.3-15.2) g/dL, white blood cells count (WBC) 2,700 (3,500-9,100) /μL, platelets 12.4 (13.0-36.9)× 10^4/μL, immunoglobulin (Ig) G 2,113 (870-1,700) mg/dL, IgA 331 (110-410) mg/dL, IgM 334 (46-260) mg/dL, and antinuclear antibody (ANA) 1,280 (<40). A liver biopsy showed moderate inflammation and severe fibrosis (F3, A2) according to the new classification of Desmet et al (1) without plasma cell infiltration (Figs. 1a, 1b). Autoimmune hepatitis was ruled out. During the therapy, serum HCV-RNA remained positive and liver functions such as AST and ALT did not reach normal levels. After completing the 48-week course of PEG-IFN/RBV therapy in June 2007, the patient was put on PEG-IFN α-2a (90 μg) without ribavirin; however, liver functions were not normalized. During both treatment protocols, the number of platelets remained between 4 and 14×10^4/μL.

At the 30th month (October 2008), the platelet count rap-

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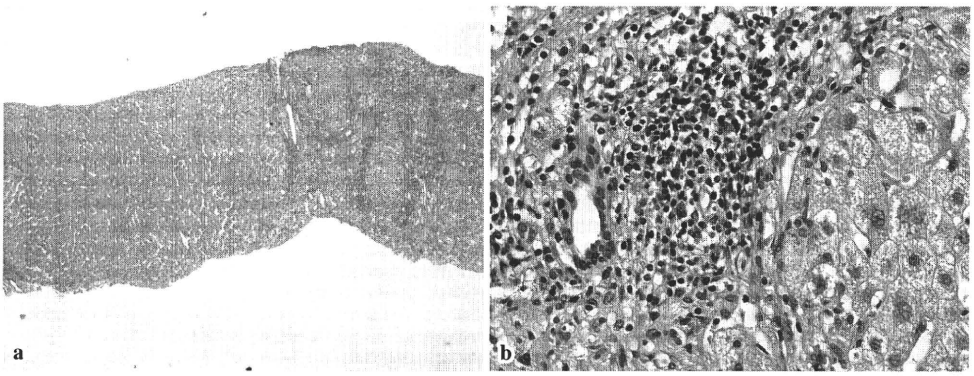

Figure 1. **Histological features of the liver: moderate inflammation and severe fibrosis (F3, A2) without plasma cell infiltration. a: Hematoxylin and Eosin staining ×40. b: Hematoxylin and Eosin staining ×400.**

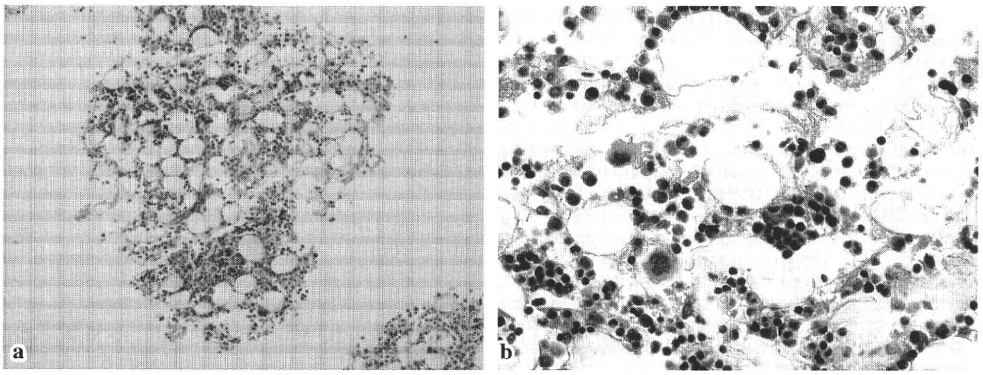

Figure 2. **Histological features of the bone marrow: normocellular marrow with myeloid: erythroid (M:E) ratio 3:1 and increased numbers of megakaryocytes (64/mm^3) compatible with a diagnosis of autoimmune thrombocytopenic purpura (AITP). a: Hematoxylin and Eosin staining ×40. b: Hematoxylin and Eosin staining ×400.**

idly declined to 1.1×10^4/μL (just 1 week before 6.2×10^4/μL), other values were WBC 1,500/μL, hemoglobin 9.0 g/dL, and hematocrit 26.8% and petechiae appeared on the patient's upper extremities. PEG-IFN α-2a was discontinued, and bone marrow biopsy showed normocellular marrow with a myeloid : erythroid (M : E) ratio of 3:1 and an increased number of megakaryocytes (64/mm^3) compatible with the diagnosis of AITP (Figs. 2a, 2b). Coagulation test results were normal, a direct Coombs' test was negative, ANA was 1,280 times (cytoplasmic×160 times), the anticardiolipin antibody was negative and cryoglobulins were positive. The platelet-associated IgG (PAIgG) level on the platelet surface had increased to 500 (normal, 9.0-25.0) ng/10^7 cells as measured by enzyme-linked immunoassay. The results of a ^{13}C-urea breath test were negative for *Helicobacter pylori* infection, obviating bacteria removal therapy; instead, corticosteroid pulse therapy was started with the intravenous administration of 1,000 mg methylprednisolone sodium succinate for three days, followed by 30 mg of oral prednisolone for 2 weeks and gradually reduced to 5 mg per day. The platelet count reverted to 6.4×10^4/μL in 14 days, and remained normal while the prednisolone dose was tapered off until the 39th month (July 2009). The PAIgG titer decreased to 57 ng/10^7 cells in response to the corticosteroid therapy (Fig. 3). HCV-RNA remained positive and liver functions did not return to normal after the withdrawal of PEG-IFN α-2a.

Discussion

Mild-to-moderate thrombocytopenia is a common adverse event of treatment with conventional interferon or with PEG-IFN α, attributed primarily to bone marrow suppression, in patients with chronic hepatitis C. Nevertheless, severe, life-threatening AITP has rarely been associated with IFN treatment (2-8). The pathogenesis of AITP is not fully understood, but IgG-type antibodies against platelet membrane glycoproteins (IIb/IIIa, Ib/IX, etc.) are known to be involved (9).

AITP, an autoimmune disorder characterized by peripheral consumption of platelets and clinical manifestations of hemorrhagic diathesis (9), is a diagnosis of exclusion and often

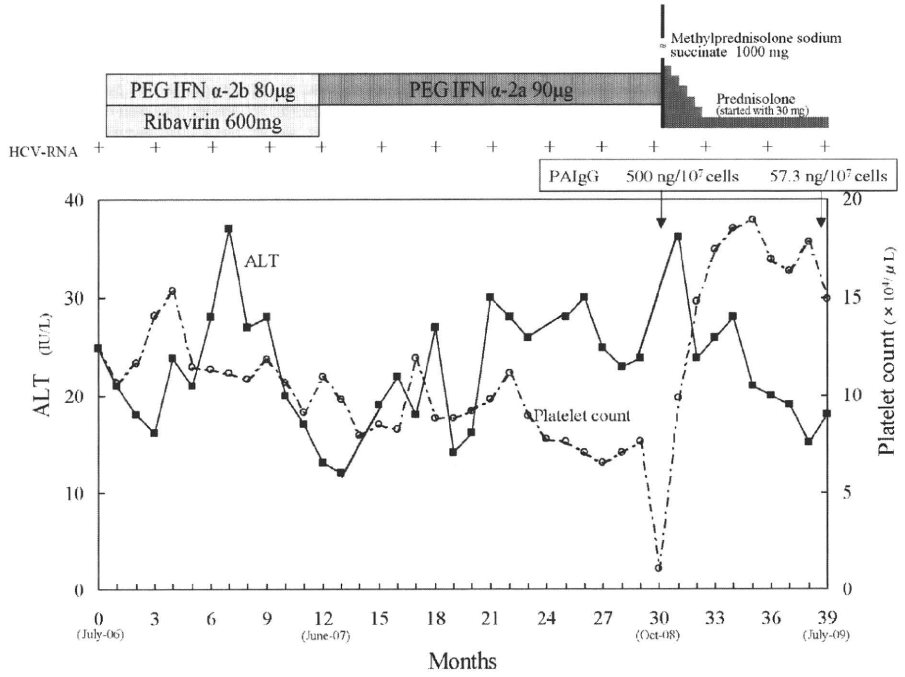

Figure 3. Clinical course of the present case.

difficult to establish. IFN-induced AITP has been reported to develop after 4 weeks to 12 months of therapy (3, 5) and even 6 months after the completion of therapy (7). AITP has been reported irrespective of the kind of IFN: IFN α-2b, PEG-IFN α-2a, and PEG-IFN with or without RBV (10).

The age of patients and baseline platelet count have varied widely, ranging from 27 (7) to 73 (6) years and from 8 (4) to 26 (5)×10^4/μL, respectively. The detection of circulating antiplatelet antibodies unbound to platelets is not sensitive enough for the diagnosis. Such autoantibodies can develop in patients immunized by pregnancy, allogenic transfusions or organ transplantation and are, thus, not specific for AITP. In contrast, direct assay of PAIgG is more useful in the diagnosis of AITP, with a sensitivity of 49-66% and a specificity of 78-92% (9). In the present case, PAIgG increased to 500 ng/10^7 cells (well above the normal range in the diagnosis of AITP) then decreased to 57.3 ng/10^7 cells (within the normal range) at the remission stage of AITP. As demonstrated in our patient, the response to steroid treatment was consistent with the diagnosis of AITP and PAIgG was also helpful in monitoring the response to corticosteroid therapy, and immunological disorders such as high γ-globulin levels of IgG and ANA positivity were found at the start of PEG-IFN/RBV therapy. Autoimmune hepatitis was ruled out by histological examination and the patient was started on PEG-IFN/RBV therapy. The clinical course was carefully monitored, focusing on the occurrence of autoimmune disease including diabetes, arthritis, sicca syndrome, vasculitis, thyroid abnormalities and others. Thirty months after the start of IFN therapy with PEG-IFN α-2b/RBV and PEG-IFN α-2a, AITP might have occurred by an autoimmunological mechanism. Clinicians should be vigilant about the occurrence of AITP during and after IFN therapy, especially in the presence of immunological disorders (11).

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Hepatology Research

Hepatology Research 2010; 40: 667–685 doi: 10.1111/j.1872-034X.2010.00673.x

Special Report

Management of hepatocellular carcinoma: Report of Consensus Meeting in the 45th Annual Meeting of the Japan Society of Hepatology (2009)

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Hepatocellular carcinoma (HCC) is responsible for approximately 600 000–700 000 deaths worldwide. It is highly prevalent in the Asia–Pacific region and Africa, and is increasing in Western countries. The evidence-based guideline for HCC in Japan was published in 2005 and revised in 2009. Apart from this guideline, a consensus-based practice manual proposed by the HCC expert panel of the Japan Society of Hepatology (JSH), which reflects widely accepted daily practice in Japan, was published in 2007. At the occasion of the 45th Annual meeting of the JSH in Kobe 4–5 June 2009, a consensus meeting of HCC was held. Consensus statements were created based on 67% agreement of 200 expert members.This article describes the up-to-date consensus statements which largely reflect the real world HCC practice in Japan. We believe readers of this article will gain the newest knowledge and deep insight on the management of HCC proposed by consensus of the HCC expert members of JSH.

Key words: hepatocellular carcinoma, Japan Society of Hepatology, staging system, surveillance, treatment algorithm, consensus-based guideline

INTRODUCTION

THE LAST EVIDENCE-BASED guideline for hepatocellular carcinoma (HCC) for Japan was published in 2005,[1] and has prevailed nationwide. This document was developed by a committee composed of 14 experts (Chairman: Professor Masatoshi Makuuchi) and was based on a critical review of 7118 English reports published between 1966 and 2002. This guideline includes 58 research questions regarding important issues for the prevention, diagnosis, surveillance and treatment of HCC. The utility of this guideline is recognized by many Japanese clinicians and has provided a great contribution to clinical practice. However, there are several issues in which solid evidence is still lacking; thus, clear recommendations for clinical practice cannot be stated. In fact, 45% of the research questions are of grade C recommendation level, representing a lack of adequate evidence. These issues are left to the clinician's discretion within the clinical setting. Furthermore, because the guidelines did not include the most up-to-date articles, no recommendation or statements were made regarding newly established evidence. In addition, the clinical practices that follow these guidelines are considered to account for 70–80% of general practice institutions.

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Received 23 December 2009; revision 3 March 2010; accepted 15 March 2010.

As mentioned above, Congress President, Professor Masatoshi Kudo, at the 45th Annual Meeting of the Japan Society of Hepatology organized the Consensus Meeting of Hepatocellular Carcinoma. The program was chaired by Professors M. Sata and S. Arii and covered the updated problems and clarified some controversial issues. Eight experts were selected to contribute to the meeting and they were assigned the following topics based on their specialties. Professor M. Sakamoto presented recommendations regarding diagnostic problems for small-sized HCC from the clinicopathological point of view. Professor M. Shimada discussed the utility of clinical staging and prognosis. Dr T. Kumada reviewed the current status of diagnostic imaging and tumor markers. Dr S. Shiina discussed important issues on ablative treatment. Dr Yamashita reviewed transarterial chemoembolization and chemotherapy. Professor N. Kokudo discussed surgical treatment, including liver transplantation. Dr M. Tanaka presented a treatment algorithm from the point-of-view of hepatologists. Finally, Professor T. Takayama comprehensively discussed the appropriateness of the present treatment algorithm.

In each presentation, the speakers raised clinical questions regarding the remaining problems that needed to be clarified in the present guidelines, and the HCC specialists (a total of 200 physicians: hepatologists, 70%; surgeons, 24%; radiologists, 2%; and pathologists, 4%) answered these questions using a question and answer analyzer system. Recommendations were approved when at least 67% of the HCC experts reached agreement. For instances where agreement was between 50% and 67%, the statements were considered informative, and are cited here as "informative statements".

In this consensus paper, each presenter has provided a summary of the recommendations and consensus. It is highly expected that this Consensus Statement established by the Japan Society of Hepatology (JSH)will provide valuable insight, and will greatly contribute to the future improvement of the guidelines and appropriate clinical practices for patients with HCC worldwide.

PATHOLOGICAL ASSESSMENT

PATHOLOGICAL ASSESSMENT OF HCC is described in the General Rules for the Clinical and Pathological Study of Primary Liver Cancer.[2] It focuses on macroscopic typing and tumor grading based on tumor differentiation and reflects the aggressiveness of the tumors; differential diagnosis between multicentric development and intrahepatic metastasis of multiple tumors; and diagnosis of early HCC and precancerous lesions. Historically, careful and detailed histological evaluation of surgical specimens enabled us to understand the clinicopathological features of HCC development and extension, and to establish the above-mentioned diagnostic criteria. However, the recent increase in non-surgical treatments for HCC, such as radiofrequency ablation (RFA), is rapidly changing the role and position of pathological diagnosis. Thus, we discussed the indications for liver tumor biopsy for the diagnosis and treatment of HCC.

When we consider the indications for liver biopsy, the risk and benefit of this procedure must be considered.[3–8] The risk includes complications caused by the procedure itself, such as hemorrhage by needle insertion, and by tumor seeding. The incidence of tumor seeding has been reported in approximately 1–5% of cases. Certainly, we have to note that the incidence depends on the characteristics of the tumor such as tumor size and tumor differentiation. Liver biopsy is important in terms of tumor diagnosis, assessment of prognosis and decision making for treatment. For example, for a typical HCC larger than 2 cm in size with a typical vascular pattern on imaging, and elevated tumor markers such as α-fetoprotein (AFP) and/or des-γ-carboxy prothrombin (DCP), the benefit of performing tumor biopsy to confirm the diagnosis of HCC seems minimal. In contrast, only liver biopsy can be used to confirm the diagnosis of cancer in cases with suspected HCC or borderline lesions on clinical and imaging diagnosis. However, controversy remains because of the inconsistent treatment strategy for suspected lesions, particularly in cases with poor liver function.

Previous follow-up data of suspected HCC and borderline lesions showed that the tumors grow slowly during the precancerous or early HCC stages, but grow rapidly in some early HCC cases or in progressed HCC.[9] The transition from slow growing to rapidly growing tumors was supposed to take place once the tumor reaches approximately 1.5 cm in size. Therefore, the proposed recommendations for liver biopsy are as follows.

Recommendation 1. Liver biopsy should be discouraged in cases with a typical HCC over 1.5 cm in size, which shows typical pattern on imaging.

Recommendation 2. Liver biopsy should be considered in cases with a suspected HCC or borderline lesions/early HCC of 1.5 cm in size or less, which does not show typical pattern on imaging.

In addition to these recommendations, the requirement of liver biopsy should increase if the detection and diagnostic ability of imaging techniques increases for

smaller lesions. The emergence of new contrast agents such as gadolinium ethoxybenzyl diethylenetriamine pentaacetic acid (Gd-EOB-DTPA) are expected to reveal suspected HCC nodules, including early HCC at approximately 1 cm in size. Tumor biopsy should then be performed to confirm the diagnosis of early cancer before it can progress to overt HCC. It is also expected that the increase in therapeutic options will increase the need for more detailed information of the tumor characteristics, such as tumor differentiation and immunophenotype reflecting tumor aggressiveness, which can only be determined by tumor biopsy.

PROGNOSTIC STAGING SYSTEM

IN TERMS OF estimating the prognosis of HCC, there are currently insufficient evidence-based data; therefore, no definite recommendations can be made, unlike other fields of HCC management. It is well known that the prognosis of HCC is defined by the behavior of the HCC itself, and by host factors such as hepatic functional reserve. The major questions that still need to be answered in terms of estimating the prognosis of HCC are: (i) whether an integrated staging system is necessary for the management of HCC; (ii) what is the best integrated staging system; and (iii) should the integrated staging system be included in the algorithm for HCC treatment?

Tumor staging (TNM staging)

There are two major classifications used for tumor staging of HCC. One is the tumor–node–metastasis (TNM) stage, developed by the American Joint Committee on Cancer (AJCC). This classification can also be applied to liver transplant recipients. However, the cut-off value for tumor diameter of 5 cm is too large to define small HCC, which are frequently found in Japan.

The other is the TNM stage proposed by the Liver Cancer Study Group of Japan (LCSGJ). The cut-off of 2 cm is very appropriate for patients in countries such as Japan, where small HCC are often found in an established nationwide screening system. However, in this system, the weighting of the strongest prognostic factor, vascular invasion, is equal to that of other factors used to estimate prognosis, which might not be adequate.

Staging for hepatic functional reserve

There are two major classifications for estimating liver functional reserve. One is the Child–Pugh classification, which is widely used worldwide, but is difficult to apply for decision making for hepatectomy. The other is the Liver Damage Classification scheme proposed by the LCSGJ, which is useful for hepatectomy. However, this scheme is not widely accepted because of the need to perform the indocyanine green retention at 15 min test ($ICGR_{15}$).

Integrated staging system for HCC

The combined classification of TNM stage and liver function stage, namely, an integrated staging system, is extremely important to estimate patient prognosis and guide decision making for patient management. The integrated staging system contributes to: (i) estimate patient prognosis; (ii) select the best treatment option for each patient; (iii) compare different treatment modalities; and (iv) compare treatment outcomes among different institutions.

Since the Okuda classification in 1985,[10] several integrated staging systems have been reported, including the Cancer of the Liver Italian Program (CLIP) score,[11] the Barcelona Clinic Liver Cancer (BCLC) stage[12] and the Japan Integrated Staging (JIS) score.[13] The Okuda classification scheme is simple and has been found to be suitable in the past, but does not seem to be suitable at the present time, now that relatively small HCC can be detected. The CLIP score is popular in Western countries, but its discriminating power is weak for small HCC, particularly at higher scores of 4–6, and over 50% of Japanese HCC patients are classified as score 0. The BCLC staging is thought to be useful as an integrated staging system and for guiding treatment. Therefore, it is recommended as an integrated treatment algorithm by the European Association for the Study of the Liver and the American Association for the Study of Liver Disease (AASLD). However, it is not suitable for the estimation of patient prognosis, and a large number of variables are used. In contrast, the JIS score essentially consists of the Child–Pugh score and the LCSGJ TNM stage, and is widely accepted in Japan. The discriminating power for relatively small HCC is excellent, and is particularly suitable for countries such as Japan, where many small HCC are detected.

In terms of a comparison of these integrated staging systems, Cillo *et al.*[14] reported that the BCLC was the best system among the Okuda, CLIP, BCLC and French classifications. Meanwhile, Tateishi *et al.*[15] reported that the Tokyo score was superior to BCLC staging and comparable to the CLIP score in predicting prognosis after hepatectomy and ablation. Kudo *et al.*[16] reported that the JIS score was better than the CLIP score, particularly in terms of discriminating power for each subgroup. Similarly, Chung *et al.*[17] reported that the JIS score was

the most excellent staging system among the BCLC, Tokyo and JIS staging systems. Therefore, JIS score is currently considered to be the best integrated staging system in Japan. Regarding other integrated staging systems, modified JIS score has been reported[13,18] to be useful for patients undergoing hepatectomy. Biomarker combined JIS score has also been reported to be useful in discrimination in patients with good prognosis.[19] However, the usefulness of these new staging systems will remain unclear until they are assessed in a range of patient sets with HCC.

Regarding the estimation of HCC prognosis, most hepatologists recognize the importance of an integrated staging system rather than applying the TNM stage and hepatic functional reserve scales individually. Furthermore, the JIS score is considered to be the best integrated staging system for current clinical practice. However, it is still difficult to incorporate the integrated staging systems, such as the JIS score, into algorithms for HCC treatment.

Recommendation 3. Integrated staging system should be used to assess the prognosis of patients with HCC, instead of individually applying scales for TNM stage and liver function stage.

Recommendation 4. The JIS score is the best staging system to estimate the prognosis of patients with HCC.

Informative Statement 1. Integrated staging systems, such as the JIS score, are not yet suitable for inclusion in algorithms for HCC treatment.

SURVEILLANCE AND DIAGNOSIS

Surveillance programs

IT IS WELL known that HCC mainly occurs in cases with chronic liver disease, particularly cirrhosis. Several cohort studies have shown that the surveillance of high-risk patients with hepatitis B virus (HBV)- or hepatitis C virus (HCV)-related chronic liver disease improves the rate of early detection and the rate of curative treatments.[20–27] For this reason, UK[28], European[29] and American[3] practice guidelines for HCC recommend routine surveillance of HCC among individuals with viral hepatitis or cirrhosis. Almost all gastroenterologists in Japan conduct surveillance programs using a combination of tumor markers such as AFP, the *lens culinaris* agglutinin-reactive fraction of AFP (AFP-L3%) and DCP, and by ultrasound (US).[30] However, no consensus has been reached in terms of the optimal surveillance strategy. Thompson *et al.* calculated the number of people who need to be under surveillance to prevent either a single death from HCC or a single premature death (defined as death before the age 75 years) and showed the effectiveness of surveillance programs.[31] In the absence of surveillance, approximately 20% of the mixed etiology cohort died as a result of HCC.

Recommendation 5. Surveillance with US and three tumor markers including AFP, DCP and AFP-L3 should be performed for early detection of HCC in patients with HBV- and HCV-related chronic liver disease, particularly cirrhosis.

Tumor markers

In Japan, AFP, AFP-L3 and DCP are widely and routinely used as serological tumor markers for the surveillance, diagnosis and prognostic estimation of HCC. The Evidence-Based Clinical Practice Guidelines of HCC published in 2005[1] recommended that AFP, AFP-L3 and DCP should be measured at intervals of 3–4 months for very high-risk patients (defined as HBV- or HCV-related liver cirrhosis), and at 6-month intervals for high-risk patients (defined as HBV- or HCV-related chronic liver disease or other causes of liver cirrhosis).[32] Although AFP is the most widely used tumor marker for HCC, the levels of AFP are also increased in patients with liver diseases other than HCC, including viral hepatitis, with a prevalence of 10–42%.[33–35] In contrast, AFP-L3 and DCP are very specific for HCC, compared with AFP alone. The combination assay for AFP, AFP-L3 and DCP should be performed for the early detection of HCC.[36,37] The specificity and sensitivity of the combination assay of AFP and DCP were 83% and 84%, respectively, to detect small HCC of less than 3 cm in diameter.[38] The specificity and sensitivity of the combination assay of DCP and AFP-L3 were 41.7–66.7% and 89.5–89.8%, respectively, to detect small HCC of less than 3 cm in diameter.[39,40]

Recommendation 6. Periodical measurement of more than two kinds of tumor markers (particularly AFP and DCP) is recommended for the early detection of HCC in high-risk and very high-risk patients.

Recommendation 7. The surveillance interval needs to be shorter in very high-risk patients than in high-risk patients.

Imaging modalities

Periodic follow-up of chronic liver disease by US, multidetector row computed tomography (MDCT) and magnetic resonance imaging (MRI) allows relatively