Oncologist®

Antiangiogenic Therapies for Advanced Hepatocellular Carcinoma

KEERAN R. SAMPAT, BERT O'NEIL

Lineberger Comprehensive Cancer Center, University of North Carolina, Chapel Hill, North Carolina, USA

Disclosures of potential conflicts of interest may be found at the end of this article.

Key Words. Hepatocellular carcinoma • Vascular endothelial growth factor • VEGF • FGF • Angiogenesis

ABSTRACT

Hepatocellular carcinoma (HCC) is a significant cause of death worldwide. HCC is a highly vascular tumor, and proangiogenic cytokines such as vascular endothelial growth factor (VEGF), platelet-derived growth factor (PDGF), and fibroblast growth factor may play crucial roles in this disease. Sorafenib, a multikinase inhibitor that blocks VEGF and PDGF signaling, was the first systemic therapy to demonstrate improved survival in patients with advanced HCC. Several other drugs targeting VEGF are in development. Because of the anticipation of eventual resistance to anti-VEGF therapies, drugs that also target alternative proangiogenic pathways are being investigated. Recent clinical and preclinical data along with ongoing studies are reviewed. ***The Oncologist*** 2013;18:430–438

Implications for Practice: Advanced hepatocellular carcinomas are refractory to most anticancer agents. Targeting of angiogenesis appears to be the most successful strategy to date for extending survival for patients with advanced HCC. This article discusses ongoing studies aimed at improving upon antiangiogenic therapy.

INTRODUCTION

Liver cancer is a major problem worldwide, accounting for more than 748,000 new cases and more than 695,000 deaths in 2008 [1]. The highest liver cancer rates are found in East and Southeast Asia. However, Western countries have seen an increase in the incidence of liver cancer in recent decades, likely due to increases in hepatitis C virus (HCV) infection and obesity—two of the most prominent risk factors for hepatocellular carcinoma (HCC) [2]. HCC accounts for 85%–90% of primary liver cancers. Most patients with HCC present with advanced-stage disease and consequently are not candidates for curative treatments, such as liver transplantation or surgical resection [3]. Even patients who undergo resection are likely to have recurrent disease, particularly those with larger tumors or tumors displaying vascular invasion [4]. Therefore, there is a clear need for development of additional agents in the management of this disease.

HCC is usually a highly vascular tumor, thus providing an attractive target for the development of new anticancer drugs. Tumor angiogenesis is a complex process that is regulated by many factors (Fig. 1) [5]. Vascular endothelial growth factor (VEGF), platelet-derived growth factor (PDGF), and basic fibroblast growth factor (FGF) all appear to play important roles in angiogenesis in many cancers, including HCC [6]. For instance, VEGF and its receptors 1, 2, and 3 have been found to be overexpressed relative to normal liver tissue in HCC, and overexpression of VEGF has been correlated with poor prognosis [7, 8]. VEGF expression was also found to increase gradually during multistep hepatocarcinogenesis [9]. PDGF is required for the recruitment of smooth muscle cells, or pericytes, that surround new blood vessels [10]. Pericytes are considered to be supportive cells for endothelial cells; they play important roles in arteriogenesis, modulation of blood flow, and regulation of vascular permeability. Basic FGF stimulates endothelial cell migration, capillary branching, and the activity of proteases, which are also essential for angiogenesis [11]. FGF has also been shown in a murine model system to act synergistically with VEGF in the progression of HCC [12] and has been implicated in escape from VEGF inhibition, as discussed herein.

SORAFENIB

Sorafenib is an oral multitargeted tyrosine kinase inhibitor (TKI) that inhibits VEGF receptors (VEGFR)-1, VEGFR-2, VEGFR-3, PDGF receptors (PDGFR)-α, PDGFR-β, c-KIT, and B-Raf, and to a lesser degree, many other kinases [13]. Sorafenib has been shown to promote apoptosis in HCC cell lines and inhibit angiogenesis in HCC xenografts [14]. Sorafenib was originally approved by the U.S. Food and Drug Administration (FDA) for treating patients with advanced-stage renal cell carcinoma; it has since been approved in many countries for treating patients with advanced HCC [13].

Approval of sorafenib in the setting of HCC was based on results of the Sorafenib Hepatocellular carcinoma Assessment Randomized Protocol (SHARP) study—a multicenter, phase III, double-blind, placebo-controlled trial that compared sorafenib to placebo in 602 patients with locally advanced or

Correspondence: Bert O'Neil, M.D., Department of Medicine, Division of Hematology/Oncology, 3rd Floor POB, CB 7305, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599, USA. Telephone: 919-966-4431; Fax: 919-966-6735; E-mail: bert_oneil@med.unc.edu Received September 26, 2012; accepted for publication January 3, 2013; first published online in ***The Oncologist*** *Express* on April 10, 2013. 1083-7159/2013/$20.00/0 http://dx.doi.org/10.1634/theoncologist.2012-0388

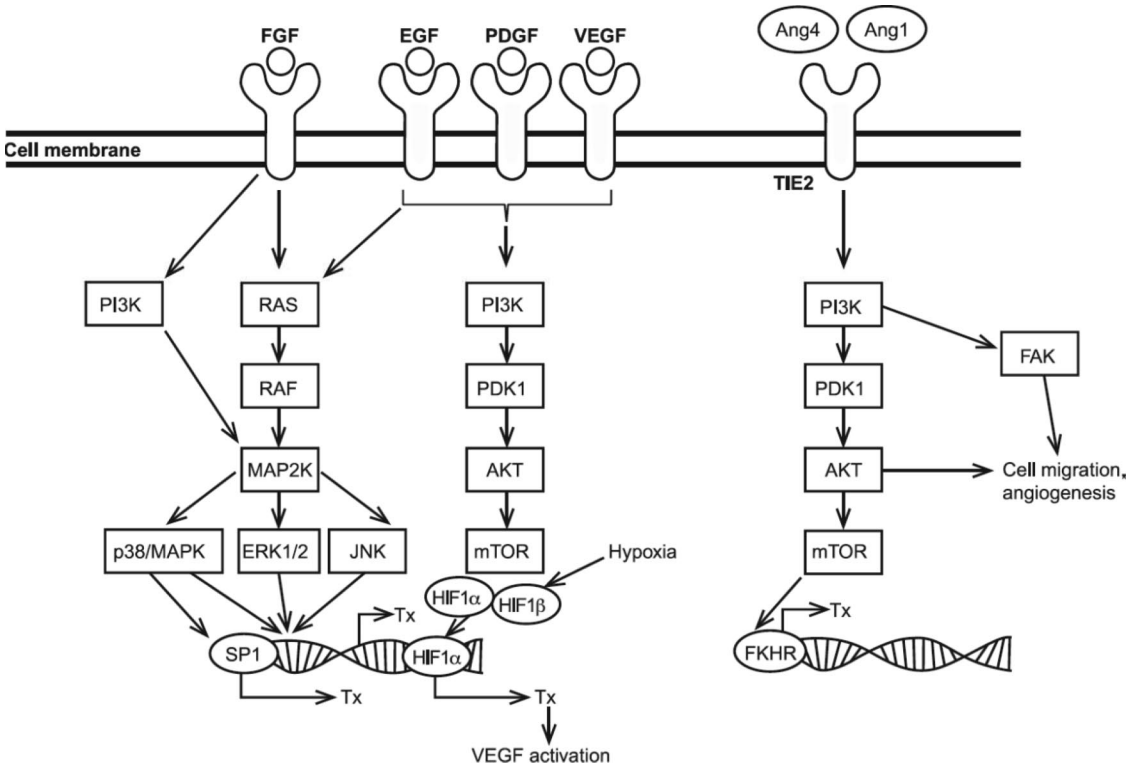

Figure 1. Alternative angiogenic signaling. All transcriptions noted can lead to angiogenesis.

Abbreviations: EGF, epidermal growth factor; FGF, fibroblast growth factor; HIF, hypoxia inducible factor; mTOR, mammalian target of rapamycin; PDGF, platelet-derived growth factor; SP1, specificity protein-1; Tx, transcription; VEGF, vascular endothelial growth factor.

metastatic HCC and predominantly Child-Pugh class A liver disease [15]. Approximately one third of the patients included in this study underwent prior embolization, but no prior systemic therapy was allowed. Median overall survival (OS) was significantly longer in the sorafenib group compared with the placebo group: 10.7 versus 7.9 months, respectively (hazard ratio [HR]: 0.69, 95% confidence interval [CI]: 0.55–0.87; $p <$ .001). Median time to progression (TTP), according to Response Evaluation Criteria in Solid Tumors, was also significantly longer in the sorafenib group compared with the placebo group: 5.5 versus 2.8 months, respectively (HR: 0.58, 95% CI: 0.45–0.74; $p <$ .001). This benefit was achieved in spite of a radiographic response rate of only 2%. The main treatment-related side effects were diarrhea (39% of patients), hand-foot syndrome (21%), fatigue (22%), and rash (16%). The magnitude of benefit of sorafenib in the SHARP trial was confirmed in another phase III study conducted in the Asia-Pacific region (NCT00492752; ClinicalTrials.gov), in which the hazard ratio was remarkably similar, but control and experimental arms had lower survival due to a preponderance of high-risk factors such as poor liver function and extrahepatic disease [16].

Because both of these randomized trials were restricted to patients with Child-Pugh A liver disease, questions have remained about use of sorafenib in patients with more advanced liver disease, for whom prognosis is known to be significantly worse [5]. The Global Investigation of Therapeutic Decisions in Hepatocellular Carcinoma and of its Treatment with Sorafenib (GIDEON) study, initiated at the time of sorafenib's approval to monitor the safety of the real-world use of sorafenib, is an ongoing global, noninterventional, prospective study of patients with advanced HCC treated with sorafenib [17]. A recent interim analysis revealed that treatment-related adverse effects were similar in patients with Child-Pugh A and B liver disease; however, a greater percentage of patients with Child-Pugh B liver dysfunction had to discontinue treatment due to adverse effects (38% vs. 23%). In the intention-to-treat population (1,614 patients), preliminary OS was 10.5 months in the Child-Pugh A group and 4.8 months in the Child-Pugh B group. This study suggests that it may be safe to use sorafenib in patients with a higher degree of liver dysfunction if close attention is paid to side effects, but median survival is very short in spite of sorafenib use. Further study is warranted before use of sorafenib in the Child-Pugh B population is considered standard.

Because many patients' tumors are initially refractory to sorafenib and all tumors eventually develop secondary resistance, there has been interest in combining sorafenib with conventional chemotherapy in an effort to improve outcomes. A phase II trial was conducted comparing doxorubicin alone versus doxorubicin plus sorafenib in patients with advanced HCC with Child-Pugh A liver dysfunction and no prior chemoembolization [18]. At the time of trial design and accrual, doxorubicin was an accepted control treatment for randomized trials in HCC. Compared with doxorubicin monotherapy, the sorafenib-doxorubicin combination was associated with significantly increased median TTP as evaluated by radiologic progression (6.4 vs. 2.8 months; $p = .02$), progression-free survival (PFS; 6.0 vs. 2.7 months; $p = .006$), and OS (13.7 vs. 6.5 months; $p = .006$). This magnitude of benefit seems greater than that seen with sorafenib alone in the SHARP study but, due to the lack of a sorafenib monotherapy

group, potential synergism between sorafenib and doxorubicin could not be confirmed. The study investigators appropriately concluded that the combination of sorafenib and doxorubicin should not yet be adopted into routine clinical use. However, the results of this trial served as the basis of an ongoing phase III Alliance (formerly Cancer and Leukemia Group B) trial comparing sorafenib plus doxorubicin with sorafenib alone (NCT01015833; ClinicalTrials.gov).

The efficacy of sorafenib in the advanced setting has led to the thought that antiangiogenic therapy could augment the efficacy of embolic therapy, particularly given findings that embolization results in transient but significant increases in serum VEGF [19]. The Sorafenib or Placebo in Combination with Transarterial Chemoembolization (SPACE) trial was a large randomized phase II trial (307 patients) that randomized patients to sorafenib 400 mg twice daily or placebo with drug-eluting bead transarterial chemoembolization (DEB-TACE) [20]. The primary endpoint was time to radiologic progression and OS was a secondary endpoint. For the sorafenib arm, the HR for TTP was 0.797 (95% CI: 0.588–1.080; $p = .072$) and the HR for OS was 0.898 (95% CI: 0.606–1.330; $p = .295$). The authors concluded that the study met its primary endpoint of improving TTP. However, the study was unusual in that a p value of .15 was considered significant based on the trial's design. Overall survival data were immature. These results do not suggest a large benefit of sorafenib in this setting, but they do support the continuation of more definitive studies, such as the ongoing Eastern Cooperative Oncology Group 1208 trial (transarterial chemoembolization with sorafenib versus transarterial chemoembolization alone; NCT01004978; ClinicalTrials.gov) in patients with liver-only disease.

Emerging Antiangiogenic Therapies for HCC

Phase III Trial of Sunitinib Versus Sorafenib

Several other drugs that target angiogenesis have been studied or are being evaluated in phase III trials for advanced HCC (Tables 1, 2). One agent to report in a phase III trial against sorafenib has been sunitinib. Sunitinib is an oral multitargeted TKI with activity against VEGFR-1, VEGFR-2, PDGFR-α, PDGFR-β, c-KIT, FLT3, and various other kinases [21]. The kinase inhibition spectrum of sunitinib is broader than sorafenib and broader than most other kinase inhibitors studied to date [22].

Based on promising phase II results, a phase III study (SUN 1170) comparing sorafenib with sunitinib was performed [23]. More than 1,000 patients with Child-Pugh A liver disease and advanced HCC were randomized between sorafenib and sunitinib. Median OS was 10.0 months for sorafenib versus 8.1 months for sunitinib (HR: 1.31, 95% CI: 1.13–1.52; $p = .0019$) [23]. As such, the study did not meet prespecified endpoints of superiority or noninferiority for sunitinib. Interestingly, PFS was 3.0 versus 3.6 months (HR: 1.13, 95% CI: 0.98–1.29; $p = .1386$) in the sorafenib and sunitinib arms, respectively. Furthermore, serious adverse events were more common in the sunitinib group than in the sorafenib group. This study was therefore stopped early due to safety concerns with sunitinib and statistical inferiority in OS of patients taking sunitinib compared with sorafenib. As such, further development of sunitinib in HCC is unlikely.

Given the activity seen with bevacizumab, it is somewhat surprising that phase III trials have not yet occurred for HCC. This may in large part be due to concerns over variceal hemorrhage. In light of this risk, the use of screening upper endoscopy along with primary preventive strategies may help minimize the risk of such fatal hemorrhages.

Summary of Phase II/III Trials

A relatively large number of phase II studies have now been conducted with alternative antiangiogenic agents. Some of these alternative therapies could hold advantages over sorafenib due to differential selectivity of drug targets or could be useful in patients who have failed sorafenib.

Bevacizumab is a recombinant humanized antibody against VEGF isoform A (which acts primarily through VEGF receptors 1 and 2) that is currently FDA-approved for treating patients with metastatic colorectal cancer and advanced nonsquamous non-small cell lung cancer, metastatic kidney cancer, and glioblastoma multiforme [24]. In a phase II study enrolling 46 patients with unresectable HCC and Child-Pugh A or B liver dysfunction and no extrahepatic metastases, bevacizumab was given intravenously (IV) every 2 weeks at a dose of 5 ($n = 12$) or 10 mg/kg ($n = 34$) until disease progression [25]. The primary objective of this study was to determine whether 6-month PFS was greater than 60% in the population treated with bevacizumab (an endpoint used because response is considered to be unlikely with angiogenesis-targeting agents). The study found that 65% of patients were progression-free at 6 months (95% CI: 51%–79%). Median PFS was 6.9 months (95% CI: 6.5–9.1 months) and median OS was an encouraging 12.4 months (95% CI: 9.4–19.9 months). Grade 3 or 4 adverse events included hypertension (15%), major bleeding (11%, including 1 fatality due to variceal bleeding), and thrombosis (6%). As a secondary endpoint, significant reductions in tumor enhancement by dynamic contrast enhanced magnetic resonance imaging were documented but did not correlate with outcome. Other phase II studies of bevacizumab have shown similar activity [26–28]. Given the activity seen with bevacizumab, it is somewhat surprising that phase III trials have not yet occurred for HCC. This may in large part be due to concerns over variceal hemorrhage. In light of this risk, the use of screening upper endoscopy along with primary preventive strategies may help minimize the risk of such fatal hemorrhages [29].

A subsequent single-arm phase II trial enrolled 40 patients with advanced HCC with Child-Pugh A and Child-Pugh B liver dysfunction to receive bevacizumab (10 mg/kg every 14 days) and daily erlotinib (150 mg), an oral epidermal growth factor receptor (EGFR) inhibitor [30]. The result for the primary endpoint of the study (PFS at 16 weeks) was 62.5%, a figure considered positive per the trial design. Median PFS was a highly encouraging 9.0 months (95% CI: 26–45 weeks) and median OS was 15.7 months (95% CI: 48–78 weeks). The major adverse effects were fatigue (20%) and hypertension (15%). Two patients did develop life-threatening gastrointestinal bleeding and one eventually died. A large randomized phase II trial

Table 1. Randomized phase III antiangiogenic therapy trials

Phase	Compound	Description and primary targets	Setting	Acronym	NCT no.	COOP	Control arm	Expected patient enrollment	Expected completion date
III	Sorafenib	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	First line		NCT00492752		Placebo	271	Mar. 2007
III	Linifanib	Multikinase inhibitor of VEGFR, PDGFR-β, CSF-1R, and others	First line		NCT01009593		Sorafenib	1,100	May 2012
III	Brivanib	Selective inhibitor of VEGFR and FGFR tyrosine kinases	First line	BRISK FL	NCT00858871		Sorafenib	1,050	Dec. 2012
III	Sorafenib + erlotinib	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); inhibitor of tyrosine kinases on EGFR	First line	SEARCH	NCT00901901		Sorafenib + placebo	731	Dec. 2012
II/III	Thalidomide+ capecitabine	Small molecule with antiangiogenic properties; 5-fluorouracil that inhibits DNA synthesis	Unspecified	OTCHCC	NCT01438450	All India Institute of Medical Science	Supportive care	74	Sept. 2014
III	Sorafenib	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	First line	BOOST (CPB status)	NCT01405573	National Cancer Institute, Naples	Supportive care	320	Mar. 2014
III	Sorafenib + cisplatin + fluorouracil	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); platinum compound that triggers apoptosis by binding to and causing DNA crosslinking; thymidylate synthase inhibitor	Unspecified	SILIUS	NCT01214343	Ministry of Health, Labor and Welfare, Japan	Sorafenib	190	Sept. 2013
III	Sorafenib + pravastatin	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); immune modulator with multiple antineoplastic effects	Unspecified		NCT01075555	FFCD-PRODIGE-11	Sorafenib	474	Sept. 2013
III	SIR-spheres		First line		NCT01135056	Singapore General Hospital	Sorafenib	360	Jul. 2015
III	SIR-spheres		First line	SARAH	NCT01482442	Assistance Publique Hopitaux de Paris P101103	Sorafenib	400	Mar. 2015
III	Sorafenib + doxorubicin	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); anthracycline antibiotic that intercalates DNA	First line		NCT01015833	CALGB-80802	Sorafenib	480	Unknown (recruiting)
III	Ramucirumab	Fully human monoclonal antibody to (receptor antagonist) against VEGFR-2	Second line	REACH	NCT01140347		Placebo	544	Apr. 2013
III	Brivanib	Selective inhibitor of VEGFR and FGFR tyrosine kinases	Second line	BRISK-PS	NCT00825955		Placebo	414	May 2012
III	Brivanib	Selective inhibitor of VEGFR and FGFR tyrosine	Second line	BRISK-APS	NCT01108705		Placebo	252	Sept. 2014
III	Sorafenib + TACE	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant		NCT01004978	ECOG-E1208	TACE + placebo	400	Sept. 2012
III	Sorafenib + doxorubicin + TACE	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); anthracycline antibiotic that intercalates DNA	Adjuvant	TACE-2	NCT01324076	CRUK-HE3005 University College London	TACE + doxorubicin	412	Nov. 2014
III	Sorafenib	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant	STORM	NCT00692770		Placebo	1,115	Oct. 2014
III	Thalidomide + TACE	Small molecule with antiangiogenic properties	Adjuvant		NCT00921531	Fudan University Liver Cancer Institute LCI-09–06-15	TACE	200	Apr. 2013
II/III	Thalidomide + RFA	Small molecule with antiangiogenic properties	Adjuvant	LDT-RFA	NCT00728078	Sun Yat-sen University	RFA	200	Unknown
III	PI-88 after surgical resection	Highly sulfonated oligosaccharide that inhibits heparanase activity and competes with heparan sulfate for FGF and VEGF binding	Adjuvant	PATRON	NCT01402908		Placebo after surgical resection	500	Dec. 2015
III	Brivanib + TACE	Selective inhibitor of VEGFR and FGFR tyrosine kinases	Adjuvant	BRISK TA	NCT00908752		TACE + placebo	870	Mar. 2015
III	Orantinib + TACE	Tyrosine kinase inhibitor of VEGFR-2, PDGFR, FGFR and c-KIT	Adjuvant	ORIENTAL	NCT01465464		TACE + placebo	880	May 2017
III	Sunitinib + TACE	Multireceptor kinase inhibitor (VEGFR, PDGFR, c-KIT, RET, csf-1R, and FLT3)	Adjuvant	SATURNE	NCT01164202	FFCD-PRODIGE-16	TACE	190	Jul. 2013

Abbreviations: COOP, Group conducting study; if blank, the study is conducted by the drug manufacturer; CPB, Child-Pugh B; CSF-1R, colony-stimulating factor 1 receptor; EGFR, epidermal growth factor receptor; FGFR, fibroblast growth factor receptor; NCT, National Clinical Trial; PDGFR, platelet-derived growth factor receptor; RET, rearranged during transfection (receptor kinase); RFA, radio frequency ablation; TACE, transcatheter arterial chemoembolization; VEGFR, vascular endothelial growth factor receptor.

of this combination versus sorafenib is currently ongoing to determine whether the regimen should proceed to a phase III trial (NCT00881751; ClinicalTrials.gov).

VEGFR-2 signaling mediates most of the known cellular processes enacted by VEGF, including the promotion of angiogenesis in tumors [31]. Signaling through VEGFR-1, in contrast, appears to be only weakly involved in mediating the angiogenic effects of VEGF [32]. No drugs that specifically inhibit VEGFR-2 have been evaluated in patients with HCC until recently. Ramucirumab is a recombinant fully human monoclonal antibody that binds the extracellular domain of VEGFR-2. In contrast to other anti-VEGF drugs, such as bevacizumab, ramucirumab binds VEGFR-2 with high affinity and specificity, preventing all VEGF ligands from binding to VEGFR-2 [33]. A phase II study of ramucirumab was conducted in 42 previously untreated patients with Child-Pugh A or Child-Pugh B liver dysfunction [34]. Seventy-six percent of these patients had extrahepatic disease and 49% had HCV infection. Ramucirumab was given IV every 2 weeks until disease progression. Median PFS was 4.2 months for Child-Pugh A patients and 2.8 months

Table 2. Randomized phase II antiangiogenic therapy trials

Phase	Compound	Description and primary targets	Setting	Acronym	NCT no.	COOP	Control arm	Expected patient enrollment	Expected completion date
II	Vandetanib	Multireceptor tyrosine kinase inhibitor of VEGFR, EGFR, and RET	First line		NCT00508001		Placebo	78	Unknown
II	Sorafenib + gemcitabine + oxaliplatin	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); nucleoside analog used as chemotherapy; DNA synthesis inhibitor	First line		NCT00941967	CCLC-GONEXT-PRODIGE-10	Sorafenib	78	Unknown
I/II	Sorafenib + bevacizumab	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); humanized monoclonal antibody against VEGF-A	First line		NCT00867321	NCGGT-N0745	Sorafenib	97	Unknown (recruiting)
II	Sorafenib + E7050	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); inhibitor of tyrosine kinases on VEGFR-2 and c-Met	First line		NCT01271504		Sorafenib	95	Dec. 2014
II	Bevacizumab + erlotinib	Humanized monoclonal antibody against VEGF-A; inhibitor of tyrosine kinases on EGFR	First line		NCT00881751	Medical University of South Carolina MUSC-101282	Sorafenib	120	Sept. 2012
II	Sorafenib + everolimus	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); inhibitor of mTOR	First line		NCT01005199	Swiss Group for CCR SWS-SASL-29	Sorafenib	106	Unknown (recruiting)
II	Dovitinib	Multikinase inhibitor that targets FGFR3, VEGFRs, FGFR1, and PDGFR	First line		NCT01232296		Sorafenib	150	Jul. 2013
II	Sorafenib + tigatuzumab	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); humanized monoclonal antibody against TRAIL-R2	First line		NCT01033240		Sorafenib	160	Mar. 2012
II	Sorafenib + mapatumumab	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); human monoclonal antibody against the TRAIL receptor	First line		NCT01258608		Sorafenib + placebo	100	Dec. 2013
II	Sorafenib + doxorubicin	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others); anthracycline antibiotic that intercalates DNA	First line	SoraDox	NCT01272557	Martin Luther Universitat Halle-Wittenberg	Sorafenib	170	Sept. 2012
II	TACE		Unspecified	STAP	NCT01480817	Seoul National University Hospital	Sorafenib	40	Sept. 2014
II	Intedanib	Small-molecule inhibitor that binds the ATP-binding domain of VEGFR 1–3, FGFR 1–3, and PDGFR-α and -β	First line		NCT01004003		Sorafenib	115	Feb. 2013
II	Intedanib	Small molecule inhibitor that binds the ATP-binding domain of VEGFR 1–3, FGFR 1–3, and PDGFR-α and -β	First line		NCT00987935		Sorafenib	142	Jul. 2012
II	Axitinib	Tyrosine kinase inhibitor of VEGFR-1, VEGFR-2, VEGFR-3, PDGFR, and cKIT	Second line		NCT01210495		Placebo	222	Jan. 2013
II	Sorafenib + TACE	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant	TACTICS	NCT01217034	Japan Liver Oncology Group JLOG 1001	Placebo + TACE	228	Sept. 2016
II	Sorafenib + RFA or TACE	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant	SORAMIC	NCT01126645	University of Madgeburg RAD85	Sorafenib	665	Feb. 2014
II	Sorafenib + TACE	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant	SPACE	NCT00855218		Placebo + TACE	307	Mar. 2012
II	Bevacizumab + TACE	Humanized monoclonal antibody against VEGF-A	Adjuvant		NCT00049322	UCLA 0206060; NCI G02–2124	Placebo + TACE	54	Jun. 2012
II	Sorafenib + laser ablation	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant	LANEX	NCT01507064	Cardarelli Hospital	Laser ablation	40	Jan. 2015
II	Sorafenib + RFA	Multikinase inhibitor (VEGFR, PDGFR, Raf, and others)	Adjuvant		NCT00813293	Beth Israel Deaconess Medical Center	Placebo + RFA	20	Unknown (recruiting)

Abbreviations: COOP, group conducting study; if blank, the study is conducted by the drug manufacturer; CSF-1R, colony-stimulating factor-1 receptor; EGFR, epidermal growth factor receptor; FGFR, fibroblast growth factor receptor; mTOR, mammalian target of rapamycin; NCI, National Cancer Institute; PDGFR, platelet-derived growth factor receptor; RET, rearranged during transfection (receptor kinase); RFA, radio frequency ablation; TACE, transcatheter arterial chemoembolization; TRAIL, tumor necrosis factor-related apoptosis inducing ligand; UCLA, University of California Los Angeles; VEGFR, vascular endothelial growth factor receptor.

for Child-Pugh B patients. The most common serious adverse events were hypertension (12%), fatigue (10%), and bleeding (7%). An ongoing phase III trial is evaluating the use of ramucirumab in patients with HCC in the second-line treatment setting after sorafenib (NCT0114034; ClinicalTrials.gov).

Linifanib is an orally available TKI with potent activity against VEGFR-1, -2, and -3 and PDGFR-β in kinase assays and a much more limited kinase inhibition spectrum than sorafenib or sunitinib [35]. In a multicenter phase II trial of linifanib with 44 patients with advanced HCC with Child-Pugh A or B liver dysfunction, median OS was 10.4 months (95% CI: 8.4–14.9) in the Child-Pugh A group ($n = 38$) and 2.5 months (95% CI: 1.1–4.5) in the Child-Pugh B group ($n = 6$) [36]. The most common serious side effects were hypertension (18%) and fatigue (14%). These results suggested that linifanib is clinically active in patients with advanced HCC and Child-Pugh A liver dysfunction. A phase III study comparing linifanib to sorafenib in patients with advanced HCC and Child-Pugh A status has completed enrollment and results are expected within the next year (NCT01009593; ClinicalTrials.gov).

Cediranib is another potent oral TKI that (relatively) specifically targets VEGFR-1 and -2 [37]. In a phase II study of daily oral cediranib enrolling 28 patients with unresectable HCC, median OS was 5.8 months (95% CI: 3.4–7.3 months) and the median TTP was 2.8 months (95% CI: 2.3–4.4 months) [37]. Twenty-six patients (93%) experienced a grade 3 or greater treatment-related adverse event; these included fatigue, anorexia, hypertension, and elevated alanine aminotransferase. Because of these toxicities, cediranib at the doses and schedule given in this trial does not

appear to be as effective or safe as other agents in the class for patients with HCC.

Brivanib is an oral, selective, dual inhibitor of FGF receptor (FGFR) and VEGFRs that has demonstrated potent antitumor and antiangiogenic effects in preclinical models of various tumor types, including HCC [38, 39]. Brivanib has also been associated with delayed tumor growth and increased survival in an HCC xenograft model of acquired resistance to sorafenib [40]. A phase II study was completed in patients with advanced HCC with Child-Pugh A and B liver status who did not receive prior systemic therapy (first-line therapy cohort; $n = 55$) [41] or who received prior treatment with at least one regimen of antiangiogenic therapy, primarily sorafenib (second-line therapy cohort; $n = 46$) [42]; 64% of these patients were Asian. In the first-line therapy cohort, median OS was 10 months (95% CI: 6.8–15.2) and median PFS was 2.7 months (95% CI: 1.4–3.0), based on modified World Health Organization tumor response criteria [41]. Interestingly, results for the second-line therapy cohort were nearly as good as those for the untreated patients, with median OS of 9.79 months (95% CI: 5.52–13.17) [42]. This suggests that brivanib may in fact help reverse one or more mechanisms of resistance to sorafenib, as will be discussed further below. The most common treatment-related side effects were elevated liver enzymes (87.3% of patients experienced elevated aspartate aminotransferase and 85.5% of patients experienced elevated alanine aminotransferase), proteinuria (65.5%), fatigue (45.5%), hypertension (45.5%), nausea (38.2%), and diarrhea (41.8%) [41].

Brivanib has been or is being investigated in several phase III trials, including first-line treatment with brivanib versus sorafenib (BRISK-FL; NCT00858871; ClinicalTrials.gov), second-line treatment with brivanib after progression on sorafenib (BRISK-PS; NCT00825955; ClinicalTrials.gov), second-line treatment with brivanib after progression on sorafenib in patients from the Asia-Pacific region (BRISK-APS; NCT01108705; ClinicalTrials.gov), and transarterial chemoembolization with or without brivanib (BRISK-TA; NCT00908752; ClinicalTrials.gov). Results from the second-line study (after sorafenib failure) have been presented in abstract form [43]. BRISK-PS randomized 395 patients in a 2:1 fashion between brivanib 800 mg/day orally and placebo. Patients entering the study were required to have been treated with sorafenib for at least 14 days and then progressed with or became intolerant to sorafenib. The primary endpoint was overall survival and the study was statistically powered to detect an HR of 0.67. The study was conducted worldwide, and patient demographics reflected a slight preponderance of Asian HCC characteristics. The results demonstrated an HR of 0.89 with a difference in median overall survival of 1.2 months favoring the brivanib arm that did not reach statistical significance. Notably, there was increased incidence of portal vein thrombosis with brivanib (31% in the brivanib arm vs. 18% in the placebo arm). In spite of the study not meeting its primary endpoint, a difference of 1.5 months and an HR of 0.56 for TTP appears to confirm that brivanib has some activity in this patient population. These results do not rule out the possibility that brivanib will meet its planned endpoints in other trials.

Phase I/II Trials

In the past, pharmaceutical companies did not sponsor trials, especially with investigational agents, in patients with liver disease and HCC. Currently, a large number of antiangiogenic agents with the potential to affect tumor growth are being explored in clinical trials, with many in earlier stages of development. Some of the most promising agents include inhibitors of the mammalian target of rapamycin (mTOR) and multiple growth factor signaling pathways.

The mTOR signaling pathway is broadly involved in the translation of proteins, including proangiogenic factors (including VEGF). Inhibition of mTOR has been explored as a potential therapeutic strategy in renal cell carcinoma, a cancer that is clearly driven by VEGF production [44, 45]. Importantly, mTOR inhibition has been effective in tumors that are refractory to VEGF pathway inhibitors.

A phase I/II study has been completed using the oral mTOR inhibitor everolimus (at a dose of 10 mg/kg/day in phase II) in 28 patients with advanced HCC and Child-Pugh A and B liver dysfunction [46]. In all, 71% of the patients in this study had received prior therapy, including sorafenib. Median OS was 8.4 months (95% CI: 3.9–21.1) and median PFS was 3.8 months (95% CI: 2.1–4.6). Fatigue, hyperglycemia, and anemia were the most common side effects associated with everolimus. There is an ongoing global, randomized, double-blinded, placebo-controlled phase III trial investigating the use of everolimus in patients after the failure of sorafenib therapy (NCT01035229; Clinicaltrials.gov).

TSU-68 is an oral TKI that targets VEGFR-2, PDGFR, and FGFR [47]. A phase I/II study was performed with TSU-68 in 35 patients with advanced HCC with Child-Pugh A and B liver dysfunction [47]. None of these patients had been treated with sorafenib and 83% were HCV positive. The median OS was 13.1 months (95% CI: 6.9–26.6), although TTP was discordant at a median of 2.1 months (95% CI: 1.2–2.9). The main treatment-related adverse events associated with TSU-68 were diarrhea, anorexia, and abdominal pain. Based on the promising activity shown in this phase I/II trial, a phase III trial is currently recruiting participants.

Resistance to Antiangiogenic Agents

The approval of sorafenib marked a major milestone in the treatment of advanced HCC and has opened the door for the development of additional antiangiogenic agents. However, antiangiogenic agents have not been as successful as initially imagined, not only in HCC but also in other solid tumors [48, 49]. Tumors typically respond initially to anti-VEGF therapies with stability (rarely shrinkage), then quickly become resistant.

Several mechanisms have been proposed to explain how cancers resist VEGF inhibition. There appears to be adaptive or evasive resistance to VEGF inhibition along with intrinsic resistance [48]. The upregulation of alternative proangiogenic signaling pathways within tumors is thought to circumvent VEGF inhibition; a well-described alternative signaling pathway involves FGF and the FGFRs [50]. In a preclinical pancreatic neuroendocrine tumor model, FGF levels were noted to be much higher in tumors treated with VEGF inhibitors when compared with untreated tumors [51]. Furthermore, VEGF inhibition with concurrent FGF blockade showed decreased tumor growth compared to VEGF inhibition alone. A clinical study of 5-FU, leucovorin, and irinotecan (FOLFIRI) plus bevacizumab in patients with metastatic colon cancer showed that FGF lev-

els were elevated in patient plasma both prior to and after disease progression [52]. Several other cytokines and angiogenic factors were also significantly elevated in patient plasma (compared to baseline) before measurable radiographic progression, including hepatocyte growth factor, placental growth factor, stromal-derived factor-1, and macrophage chemoattractant protein-3 [52]. Recently, upregulation of the FGF-8 subfamily has been shown to promote tumor cell survival in HCC [53].

Because of structural similarities in the kinase domains of several growth factor receptors, some TKIs have multiple-receptor specificity. It is therefore not surprising that ongoing clinical trials include agents such as brivanib, dovitinib, and BIBF 1120 that target both FGFR and VEGFR. These studies may provide interesting information about whether dual inhibition of FGFR and VEGFR is more effective in the front-line setting or around the time of progression.

VEGF and VEGFR-targeting therapy has represented the first real success in HCC in many years, reinvigorating research in this deadly and difficult malignancy. The success of targeting VEGF receptors now needs to be followed by better understanding of mechanisms of intrinsic and acquired resistance to these agents. As our understanding evolves, next-generation anti-VEGFR agents and agents targeting alternative proangiogenic pathways hold significant promise for the treatment of HCC.

Acquired resistance to VEGF inhibition may also be related to changes in vascular stromal cells [54]. In a mouse xenograft model of non-small cell lung adenocarcinoma, increased expression of components of the EGFR and FGFR signaling pathways were evident in the gene expression profile of the stromal cell compartment [54]. In bevacizumab-resistant tumors, pericytes expressed higher levels of EGFR than control (non-bevacizumab resistant) tumors. Furthermore, bevacizumab-resistant tumors exhibited an increase in pericyte coverage of vessels—a necessary step in angiogenesis. Targeting both the EGFR and VEGF pathways with bevacizumab and erlotinib in mouse xenografts significantly delayed the onset of therapeutic resistance compared with inhibition of either pathway alone [54]. This suggests that tumors may switch from VEGFR- to EGFR-driven angiogenesis as resistance occurs, and it may explain the efficacy of the bevacizumab/erlotinib combination mentioned earlier.

Future Directions for Antiangiogenic Therapy

Proteins other than growth factors play key roles in tumor angiogenesis and may eventually serve as therapeutic targets in the quest to prevent resistance to antiangiogenic treatment. For example, hypoxia inducible factor (HIF)-1α, a proangiogenic transcription factor upstream of VEGF and FGF, may participate in resistance to VEGF inhibition [55]. Increased tumor hypoxia due to VEGF inhibition has been shown to lead to increased expression of HIF-1α. In a non-HCC mouse xenograft model, the addition of topotecan, a potent HIF-1α inhibitor, to bevacizumab treatment led to significant reduction in tumor growth compared to bevacizumab or topotecan treatment alone [55]. Similarly, specificity protein (SP)-1 is another transcription factor that is involved in the expression of VEGF and other proangiogenic mediators [56]. Inhibition of SP-1 with mithramycin A appears to have antiangiogenic and antitumor activity in mouse pancreatic cancer xenografts [56]. These preclinical studies suggest that targeting transcription factors such as HIF-1α and SP-1 (with drugs) would be a worthwhile strategy to investigate in tumors with robust angiogenesis.

Blood vessel integrity and normalization are emerging as critical parts of understanding angiogenesis in cancer [57]. The angiopoietin (ANGPT)-TIE (tyrosine kinase with immunoglobulin-like and EGF-like domains) system is thought to be intimately involved in vascular maintenance. ANGPTs are ligands that bind to TIE-2 receptors on vascular endothelial cells [58], influencing the maintenance of existing blood vessels and maturation of new vessels. To take advantage of this interaction, several drugs targeting the ANGPTs and TIE2 are currently in development for solid tumors and may represent an additional opportunity to target tumor vasculature [58].

VEGF and VEGFR-targeting therapy has represented the first real success in HCC in many years, reinvigorating research in this deadly and difficult malignancy. The success of targeting VEGF receptors now needs to be followed by better understanding of mechanisms of intrinsic and acquired resistance to these agents. As our understanding evolves, next-generation anti-VEGFR agents and agents targeting alternative proangiogenic pathways hold significant promise for the treatment of HCC.

At present, the lack of good radiographic and biologic biomarkers remains an additional challenge for the development of effective therapies for HCC. However, using newer imaging modalities to detect early biologic changes is an active area of research. For example, a small retrospective study demonstrated that baseline positron emission tomography-computed tomography (PET-CT) scans may predict OS and PFS for patients who undergo treatment with sorafenib [59]. Imaging techniques such as computed tomography or magnetic resonance perfusion imaging and PET-CT hold promise in assessing responses in HCC and will aid the development of biologic therapies in the future [60].

Acknowledgments

We thank Cailin Moira Wilke, Ph.D., of StemScientific (funded by Bristol-Myers Squibb) for providing assistance with editing and generation of figures.

Author Contributions

Conception/Design: Keeran R. Sampat, Bert O'Neil
Collection and/or assembly of data: Keeran R. Sampat, Bert O'Neil
Manuscript writing: Keeran R. Sampat, Bert O'Neil
Final approval of manuscript: Keeran R. Sampat, Bert O'Neil

Disclosures

Bert O'Neil: Bayer, Bristol-Myers Squibb, Genentech (C/A); Bayer, Novartis, GlaxoSmithKline (RF). The other author indicated no financial relationships.

C/A: Consulting/advisory relationship; RF: Research funding; E: Employment; H: Honoraria received; OI: Ownership interests; IP: Intellectual property rights/inventor/patent holder; SAB: scientific advisory board

References

1. Jemal A, Bray F, Center MM et al. Global cancer statistics. CA Cancer J Clin 2011;61:69–90.

2. El-Serag HB, Rudolph KL. Hepatocellular carcinoma: Epidemiology and molecular carcinogenesis. Gastroenterology 2007;132:2557–2576.

3. Llovet JM, Burroughs A, Bruix J. Hepatocellular carcinoma. Lancet 2003;362:1907–1917.

4. Nathan H, Schulick RD, Choti MA et al. Predictors of survival after resection of early hepatocellular carcinoma. Ann Surg 2009;249:799–805.

5. Thomas MB, Jaffe D, Choti MM et al. Hepatocellular carcinoma: Consensus recommendations of the National Cancer Institute Clinical Trials Planning Meeting. J Clin Oncol 2010;28:3994–4005.

6. Fernández M, Semela D, Bruix J et al. Angiogenesis in liver disease. J Hepatol 2009;50:604–620.

7. Dhar DK, Naora H, Yamanoi A et al. Requisite role of VEGF receptors in angiogenesis of hepatocellular carcinoma: A comparison with angiopoietin/Tie pathway. Anticancer Res 2002;22:379–386.

8. Tseng PL, Tai MH, Huang CC et al. Overexpression of VEGF is associated with positive p53 immunostaining in hepatocellular carcinoma (HCC) and adverse outcome of HCC patients. J Surg Oncol 2008;98:349–357.

9. Park YN, Kim YB, Yang KM et al. Increased expression of vascular endothelial growth factor and angiogenesis in the early stage of multistep hepatocarcinogenesis. Arch Pathol Lab Med 2000;124:1061–1065.

10. Conway EM, Collen D, Carmeliet P. Molecular mechanisms of blood vessel growth. Cardiovasc Res 2001;49:507–521.

11. Javerzat S, Auguste P, Bikfalvi A. The role of fibroblast growth factors in vascular development. Trends Mol Med 2002;8:483–489.

12. Yoshiji H, Kuriyama S, Yoshii J et al. Synergistic effect of basic fibroblast growth factor and vascular endothelial growth factor in murine hepatocellular carcinoma. Hepatology 2002;35:834–842.

13. Nexavar [package insert]. Wayne, NJ: Bayer Healthcare Pharmaceuticals, 2011.

14. Huynh H, Ngo VC, Koong HN et al. Sorafenib and rapamycin induce growth suppression in mouse models of hepatocellular carcinoma. J Cell Mol Med 2009;13:2673–2683.

15. Llovet JM, Ricci S, Mazzaferro V et al. Sorafenib in advanced hepatocellular carcinoma. N Engl J Med 2008;359:378–390.

16. Cheng AL, Kang YK, Chen Z et al. Efficacy and safety of sorafenib in patients in the Asia-Pacific region with advanced hepatocellular carcinoma: A phase III randomised, double-blind, placebo-controlled trial. Lancet Oncol 2009;10:25–34.

17. Marrero JA, Lencioni R, Kudo M et al. Global investigation of therapeutic decisions in hepatocellular carcinoma and of its treatment with sorafenib (GIDEON) second interim analysis in more than 1,500 patients: Clinical findings in patients with liver dysfunction. J Clin Oncol 2011;29(suppl 15):4001.

18. Abou-Alfa GK, Johnson P, Knox JJ et al. Doxorubicin plus sorafenib vs doxorubicin alone in patients with advanced hepatocellular carcinoma: a randomized trial. JAMA 2010;304:2154–2160.

19. Shim JH, Park JW, Kim JH et al. Association between increment of serum VEGF level and prognosis after transcatheter arterial chemoembolization in hepatocellular carcinoma patients. Cancer Sci 2008;99:2037–2044.

20. Lencioni R, Llovet JM, Han G et al. Sorafenib or placebo in combination with transarterial chemoembolization (TACE) with doxorubicin-eluting beads (DEBDOX) for intermediate-stage hepatocellular carcinoma (HCC): Phase II, randomized, double-blind SPACE trial. ASCO Meeting Abstracts 2012;30:LBA154.

21. Mendel DB, Laird AD, Xin X et al. In vivo antitumor activity of SU11248, a novel tyrosine kinase inhibitor targeting vascular endothelial growth factor and platelet-derived growth factor receptors: Determination of a pharmacokinetic/pharmacodynamic relationship. Clin Cancer Res 2003;9:327–337.

22. Weis SM, Cheresh DA. Tumor angiogenesis: Molecular pathways and therapeutic targets. Nat Med 2011;17:1359–1370.

23. Cheng A, Kang Y, Lin D et al. Phase III trial of sunitinib (Su) versus sorafenib (So) in advanced hepatocellular carcinoma (HCC). J Clin Oncol 2011;29(suppl 15):Abstract 4000.

24. Avastin [package insert]. South San Francisco, CA: Genetech Inc 2009.

25. Siegel AB, Cohen EI, Ocean A et al. Phase II trial evaluating the clinical and biologic effects of bevacizumab in unresectable hepatocellular carcinoma. J Clin Oncol 2008;26:2992–2998.

26. Sun W, Sohal D, Haller DG et al. Phase 2 trial of bevacizumab, capecitabine, and oxaliplatin in treatment of advanced hepatocellular carcinoma. Cancer 2011;117:3187–3192.

27. Hsu CH, Yang TS, Hsu C et al. Efficacy and tolerability of bevacizumab plus capecitabine as first-line therapy in patients with advanced hepatocellular carcinoma. Br J Cancer 2010;102:981–986.

28. Zhu AX, Blaszkowsky LS, Ryan DP et al. Phase II study of gemcitabine and oxaliplatin in combination with bevacizumab in patients with advanced hepatocellular carcinoma. J Clin Oncol 2006;24:1898–1903.

29. Vlachogiannakos J, Goulis J, Patch D et al. Primary prophylaxis for portal hypertensive bleeding in cirrhosis. Aliment Pharmacol Ther 2000;14:851–860.

30. Thomas MB, Morris JS, Chadha R et al. Phase II trial of the combination of bevacizumab and erlotinib in patients who have advanced hepatocellular carcinoma. J Clin Oncol 2009;27:843–850.

31. Shalaby F, Rossant J, Yamaguchi TP et al. Failure of blood-island formation and vasculogenesis in flk-1-deficient mice. Nature 1995;376:62–66.

32. Takahashi S. Vascular endothelial growth factor (VEGF), VEGF receptors and their inhibitors for antiangiogenic tumor therapy. Biol Pharm Bull 2011;34:1785–1788.

33. Spratlin JL, Cohen RB, Eadens M et al. Phase I pharmacologic and biologic study of ramucirumab (IMC-1121B), a fully human immunoglobulin G1 monoclonal antibody targeting the vascular endothelial growth factor receptor-2. J Clin Oncol 2010;28:780–787.

34. Zhu AX, Finn RS, Mulcahy MF et al. A phase II study of ramucirumab as first-line monotherapy in patients (pts) with advanced hepatocellular carcinoma (HCC). J Clin Oncol 2010;28(suppl 15):4083.

35. Wong CI, Koh TS, Soo R et al. Phase I and biomarker study of ABT-869, a multiple receptor tyrosine kinase inhibitor, in patients with refractory solid malignancies. J Clin Oncol 2009;27:4718–4726.

36. Toh H, Chen P, Carr BI et al. Linifanib phase II trial in patients with advanced hepatocellular carcinoma (HCC). J Clin Oncol 2010;28(suppl 15):4038.

37. Alberts SR, Fitch TR, Kim GP et al. Cediranib (AZD2171) in patients with advanced hepatocellular carcinoma: A phase II north central cancer treatment group clinical trial. Am J Clin Oncol 2012;35:329–333.

38. Huynh H, Ngo VC, Fargnoli J et al. Brivanib alaninate, a dual inhibitor of vascular endothelial growth factor receptor and fibroblast growth factor receptor tyrosine kinases, induces growth inhibition in mouse models of human hepatocellular carcinoma. Clin Cancer Res 2008;14:6146–6153.

39. Bhide RS, Lombardo LJ, Hunt JT et al. The antiangiogenic activity in xenograft models of brivanib, a dual inhibitor of vascular endothelial growth factor receptor-2 and fibroblast growth factor receptor-1 kinases. Mol Cancer Ther 2010;9:369–378.

40. Tovar V, Cornella H, Villanueva A et al. FGF signaling dysregulation in HCC and role in the development of acquired resistance to anti-angiogenic therapies. Paper presented at: 62nd Annual Meeting of the American Association for the Study of Liver Diseases; November 4–8, 2011; San Francisco, CA.

41. Park JW, Finn RS, Kim JS et al. Phase II, open-label study of brivanib as first-line therapy in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2011;17:1973–1983.

42. Finn RS, Kang YK, Mulcahy M et al. Phase II, open-label study of brivanib as second-line therapy in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2012;18:2090–2098.

43. Llovet JM, Decaens T, Raoul J-L et al. Brivanib versus placebo in patients with advanced hepatocellular carcinoma (HCC) who failed or were intolerant to sorafenib: results from the phase 3 BRISK-PS study. J Hepatol 2012;56(suppl 2):S549.

44. Motzer RJ, Escudier B, Oudard S et al. Efficacy of everolimus in advanced renal cell carcinoma: A double-blind, randomised, placebo-controlled phase III trial. Lancet 2008;372:449–456.

45. Motzer RJ, Escudier B, Oudard S et al. Phase 3 trial of everolimus for metastatic renal cell carcinoma: Final results and analysis of prognostic factors. Cancer 2010;116:4256–4265.

46. Zhu AX, Abrams TA, Miksad R et al. Phase 1/2 study of everolimus in advanced hepatocellular carcinoma. Cancer 2011;117:5094–5102.

47. Kanai F, Yoshida H, Tateishi R et al. A phase I/II trial of the oral antiangiogenic agent TSU-68 in patients with advanced hepatocellular carcinoma. Cancer Chemother Pharmacol 2011;67:315–324.

48. Bergers G, Hanahan D. Modes of resistance to anti-angiogenic therapy. Nat Rev Cancer 2008;8:592–603.

49. Rini BI, Atkins MB. Resistance to targeted therapy in renal-cell carcinoma. Lancet Oncol 2009;10:992–1000.

50. Lieu C, Heymach J, Overman M et al. Beyond VEGF: Inhibition of the fibroblast growth factor pathway and antiangiogenesis. Clin Cancer Res 2011;17:6130–6139.

51. Casanovas O, Hicklin DJ, Bergers G et al. Drug resistance by evasion of antiangiogenic targeting of

VEGF signaling in late-stage pancreatic islet tumors. Cancer Cell 2005;8:299–309.

52. Kopetz S, Hoff PM, Morris JS et al. Phase II trial of infusional fluorouracil, irinotecan, and bevacizumab for metastatic colorectal cancer: Efficacy and circulating angiogenic biomarkers associated with therapeutic resistance. J Clin Oncol 2010;28: 453–459.

53. Gauglhofer C, Sagmeister S, Schrottmaier W et al. Up-regulation of the fibroblast growth factor 8 subfamily in human hepatocellular carcinoma for cell survival and neoangiogenesis. Hepatology 2011;53:854–864.

54. Cascone T, Herynk MH, Xu L et al. Upregulated stromal EGFR and vascular remodeling in mouse xenograft models of angiogenesis inhibitor-resistant human lung adenocarcinoma. J Clin Invest 2011;121:1313–1328.

55. Rapisarda A, Hollingshead M, Uranchimeg B et al. Increased antitumor activity of bevacizumab in combination with hypoxia inducible factor-1 inhibition. Mol Cancer Ther 2009;8:1867–1877.

56. Yuan P, Wang L, Wei D et al. Therapeutic inhibition of Sp1 expression in growing tumors by mithramycin a correlates directly with potent antiangiogenic effects on human pancreatic cancer. Cancer 2007;110:2682–2690.

57. Carmeliet P, Jain RK. Principles and mechanisms of vessel normalization for cancer and other angiogenic diseases. Nat Rev Drug Discov 2011;10: 417–427.

58. Huang H, Bhat A, Woodnutt G et al. Targeting the ANGPT-TIE2 pathway in malignancy. Nat Rev Cancer 2010;10:575–585.

59. Lee JH, Park JY, Kim do Y et al. Prognostic value of 18F-FDG PET for hepatocellular carcinoma patients treated with sorafenib. Liver Int 2011;31: 1144–1149.

60. Jiang T, Zhu AX, Sahani DV. Established and novel imaging biomarkers for assessing response to therapy in hepatocellular carcinoma. J Hepatol 2012;58:169–177.