Journal of Hepatocellular Carcinoma

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REVIEW

Regorafenib Combined with Other Systemic Therapies: Exploring Promising Therapeutic Combinations in HCC

Alessandro Granito[1,2]
Sara Marinelli[1]
Antonella Forgione[1,2]
Matteo Renzulli[3]
Francesca Benevento[2]
Fabio Piscaglia[1,2]
Francesco Tovoli[1,2]

[1]Division of Internal Medicine, Hepatobiliary and Immunoallergic Diseases, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna, Italy; [2]Department of Medical and Surgical Sciences, University of Bologna, Bologna, Italy; [3]Radiology Unit, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna, Italy

Abstract: Regorafenib was the first drug to demonstrate a survival benefit as a second-line agent after sorafenib failure in patients with unresectable hepatocellular carcinoma (HCC). Recent studies have shown that its mechanism of action is not only limited to its very broad spectrum of inhibition of angiogenesis, tumor proliferation, spread, and metastasis, but also to its immunomodulatory properties that have favorable effects on the very intricate role that the tumor microenvironment plays in carcinogenesis and tumor growth. In this review, we discuss rationale and evidence supporting regorafenib efficacy in HCC and that led to its approval as a second-line treatment, after sorafenib failure. We also discuss the evidence from clinical practice studies that confirm the results previously achieved in clinical trials. Finally, we analyze the potential role of regorafenib in emerging combined treatment approach with immunotherapy strategies using immune checkpoint blockade and its potential extension to patient categories not included in the registrative study.

Keywords: hepatocellular carcinoma, HCC, regorafenib, tyrosine kinase inhibitor, TKI, systemic treatment, combination treatment

Introduction

Hepatocellular carcinoma (HCC) is the fifth most common cancer and the third most frequent cause of cancer death worldwide, with 905.677 new cases and 830.180 deaths in 2020 which are responsible for 8.3% of all cancers.[1]

Hepatocellular carcinoma (HCC) accounts for nearly 90% of primary liver cancers and is a leading world health problem. The incidence of HCC increases dramatically with age in all populations, achieving a peak at age 70, and is increasing in most countries being the dominant cause of mortality in cirrhotic patients.[2–4]

Globally, chronic viral hepatitis and alcoholic liver disease are the leading risk factors for HCC development, although in high-income areas non-alcoholic fatty liver disease (NAFLD) linked to HCC is increasing due to the increasing prevalence of metabolic disorders.[5–7]

In contrast, vaccination and treatment for hepatitis B virus (HBV) infection, prevention campaigns for sexual and iatrogenic transmission of HBV and hepatitis C virus (HCV), and the introduction of effective HCV antiviral agents are reducing the burden of chronic viral liver disease.[8–11]

Since 2007, for HCC patients with preserved liver function and advanced or intermediate Barcelona Clinic Liver Cancer (BCLC) stage, unsuitable for

Correspondence: Alessandro Granito
IRCCS Azienda Ospedaliero-Universitaria di Bologna, via Albertoni 15, Bologna, Italy
Email alessandro.granito@unibo.it

Received: 31 December 2020
Accepted: 25 March 2021
Published: 26 May 2021

locoregional treatment, the multikinase inhibitor (MKI) sorafenib has been considered the standard of care worldwide.[12] After a decade of unsatisfactory results, other agents have been approved as a first-line alternative to sorafenib, or in a second-line setting, after sorafenib failure.[13–17] Besides, immune checkpoint inhibitors (ICIs) targeting the programmed cell death receptor-1 (PD-1), anti-programmed death-ligand 1 (PD-L1), and anti-cytotoxic T lymphocyte associated antigen 4 (CTLA-4) have recently received accelerated approval.[14,18–20]

In 2016 the randomized, placebo-controlled, Phase III RESORCE trial was the first demonstrating that systemic treatment with regorafenib in patients experiencing failure of first-line therapy with sorafenib resulted in a significant increase in OS in the treatment arm compared to the placebo arm [10.6 versus 7.8 months, HR 0.63 (95% CI 0.50–0.79), p<0.0001], after a decade of failed clinical trials investigating a wide range of drugs tested for second-line treatment.[15]

Since registration and the start of its use in clinical practice, real-life experiences have also been reported.[21] Also, recent developments in the systemic treatment of HCC have opened new possible scenarios in the potential use of regorafenib in combination with other agents or new options for its sequential use.[22,23]

In this review, we examine the main preclinical and clinical results of studies evaluating regorafenib for the treatment of HCC patients and discuss the rationale for its possible use in combination treatment with other agents as well as potential options in a sequential treatment strategy.

Mechanism of Action

Regorafenib (chemical name 4-(4-(3-(4-Chloro- 3-(trifluoromethyl) phenyl) ureido)- 3-fluorophenoxy)-N-methylpicolinamide) is a small molecule inhibitor and belongs to the group of biaryl urea compounds. Regorafenib is an orally available multitargeted tyrosine kinase inhibitor (TKI) that was developed following a discovery program aimed at the optimization of the potency of sorafenib, from whom it differs only by the addition of a fluorine atom in the center phenyl ring (Figure 1).[24]

TKIs are a class of agents involved in the activation of a broad range of proteins via phosphorylation. TKIs bind to the active site of tyrosine kinases, thereby hindering phosphorylation and inhibiting downstream signal transduction of a variety of growth factors. By blocking key tyrosine kinase pathways in tumors, such as the vascular endothelial growth factor receptor (VEGFR), platelet-derived growth

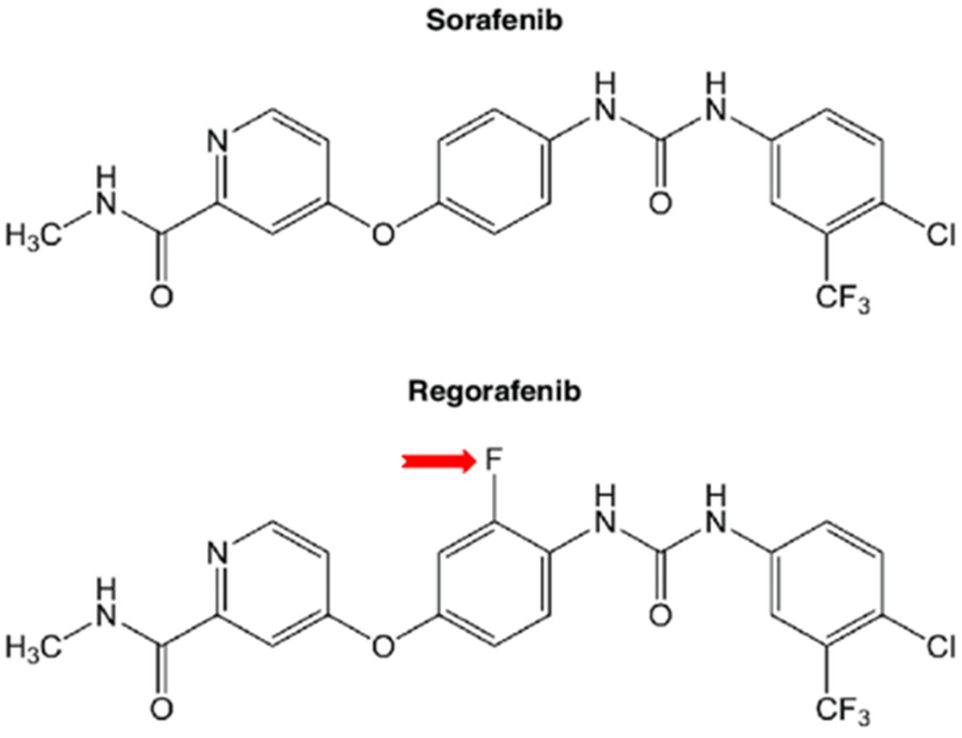

Figure 1 Similarly to sorafenib, regorafenib is a bi-aryl urea class of drug. The sole difference between sorafenib and regorafenib is the presence of a fluorine atom (red arrow) in the latter. Due to a mechanism that has not yet been fully defined, this one unique difference produces a wider kinase inhibitory profile. In complement to the targets that are inhibited by sorafenib, regorafenib also blocks the signaling pathway of Tie2, the receptor for angiopoietin-2, a pro-angiogenic cytokine.

factor (PDGFR), and epidermal growth factor receptor 2 (EGFR), they inhibit tumor growth. TKIs differ in their spectrum of inhibition, which can also simultaneously hit multiple targets and thus inhibit tumorigenesis differently.[25]

Preclinical studies have shown that regorafenib targets kinases involved in signaling pathways driving tumorigenesis, cancer progression, and tumor microenvironment maintenance. These kinases are isoforms of rapidly accelerated fibrosarcoma (RAF) RAF-1, B-RAF, B-RAFV600E (a mutant B-RAF isoform), VEGFR 1, 2 and 3, the oncogenic kinases KIT and RET, angiopoietin 1 receptor (TIE2), platelet-derived growth factor receptor (PDGFR), fibroblast growth receptors (FGFRs) 1 and 2 (Figure 2).[24,26–31]

Thus, the dual blockade of VEGF receptors and TIE2 can lead to an obvious and unique enhancement of the effect of tumor vessel reduction.

Among the approved systemic therapies for HCC with anti-angiogenic effects, regorafenib blocks a broader range of targets (Table 1).

There are also findings suggesting regorafenib's anti-immunosuppressive property and promotion of anti-tumor immunity.[32] Regorafenib has the important effect of enhancing anti-tumor immunity via macrophage modulation and increase proliferation and activation of CD8+ T cells (Figure 2). Tumor-associated macrophages (TAMs), a key element of leukocyte infiltration, enhance tumor cell growth, development and migration.[32,33] The role of TAMs in carcinogenesis is well-documented in several tumor types, including HCC.[34,35]

Regorafenib also impairs tumor immunity by inhibiting the colony-stimulating factor 1 receptor CSF1R which is critical for macrophage differentiation and survival and causes a reduction in tumor infiltration of macrophages.[29,31,36]

In agreement, regorafenib has been shown to decrease the infiltration of TAM, which is crucial for angiogenesis and metastatic spreading, and reverts their polarization from M2 pro-tumor phenotype to M1 tumor growth-inhibiting phenotype.[29,32]

Recently, the synergistic relationship between regorafenib and natural killer (NK) cells has been reported. The binding of NKG2D receptors on the surface of NK cells and NKG2DL expressed in tumor cells leads to the activation of NK cells elimination of tumor cells. However, tumor cells utilize various mechanisms to evade the NKG2D receptor/NKG2DL mediated immune clearance.[37] Tai and colleagues demonstrated that Regorafenib induces STAT3 signaling pathway inhibition, resulting in enhanced NK cell cytolytic activity via upregulation of the NKG2D ligand and assisting recognition of HCC cells by NK cells and ultimately HCC cell apoptosis.[38]

Lastly, it has been reported that long-term regorafenib therapy has been also demonstrated to lower angiogenesis and also to be beneficial to portal hypertension, and acute administration improves portal hemodynamics, indicating

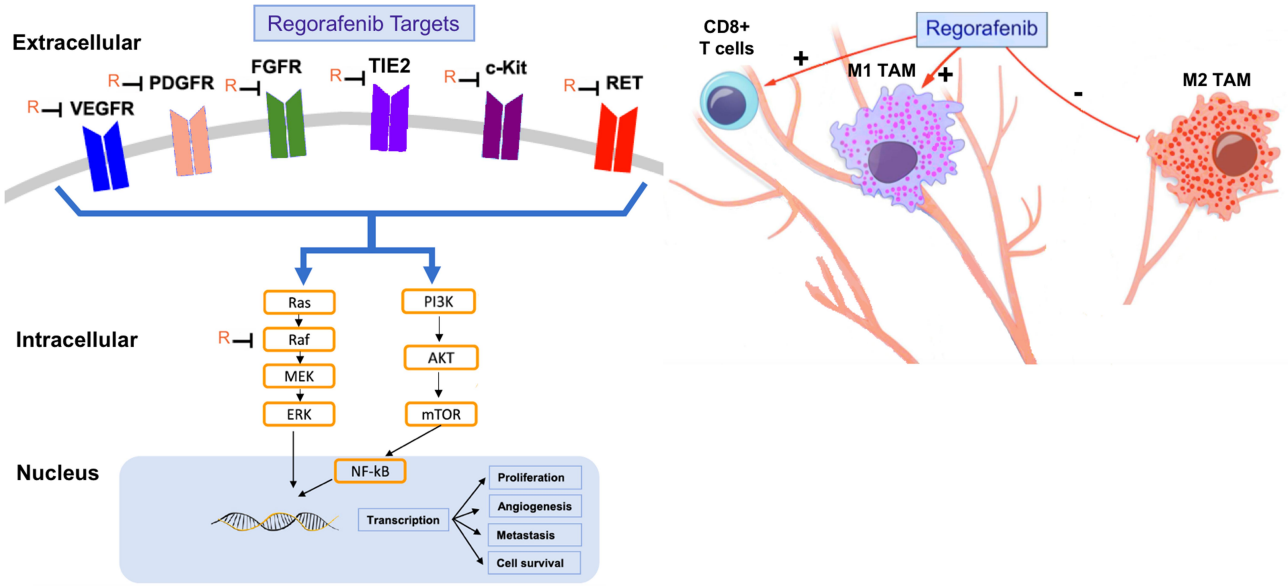

Figure 2 Regorafenib is able to inhibit several molecular pathways by targeting angiogenic, stromal, oncogenic and intracellular kinases. Regorafenib induces M1 macrophage polarization and increases CD8+ T cells proliferation and activation thus also acting on the tumor microenvironment and immunosuppression.

Table 1 Target Structures of Systemic Therapies with Antiangiogenic Effects

Line	Drug	Drug Category	Cellular Targets										
			VEGFR1	VEGFR2	VEGFR3	PDGFR	RAF	FGFR	KIT	RET	TIE-2	MET	AXL
1st	Sorafenib	TKI	♦	♦	♦	♦	♦		♦	♦			
1st	Lenvatinib	TKI	♦	♦	♦	♦		♦	♦	♦			
2nd	**Regorafenib**	**TKI**	♦	♦	♦	♦	♦	♦	♦	♦	♦		
2nd	Cabozantinib	TKI		♦					♦	♦	♦	♦	♦
2nd	Ramucirumab	TKI		♦									

Note: Among the approved systemic therapies for HCC with anti-angiogenic effects, regorafenib displays the broadest spectrum of inhibited target receptors.

that it may be particularly beneficial to patients with portal hypertension and maintained hepatic function.[39]

As such, regorafenib's broad spectrum of kinase inhibition, together with its immunomodulatory effects, may explain its established and emerging clinical activity in various tumor types and have offered a rationale for supporting clinical trials to investigate the development of a combination strategy with immune checkpoint inhibitors.[40–42]

Regorafenib is metabolized by UGT1A9 and CYP3A4 enzymes to two active metabolites M-5 (demethylated N-oxide) and M-2 (N-oxide).[43]

CYP enzymes may be inhibited or induced by the co-administration of agents that interact with the same enzymes. Co-administration of regorafenib with a strong CYP3A4 inhibitor might increase the regorafenib serum area under the curve (AUC). This could result in a potential enhancement of drug toxicity. On the contrary, co-administration of regorafenib with a significant CYP3A4 inducer would lead to a regorafenib decline in the serum AUC levels and a potential reduction in efficacy. The M-5 and M-2 regorafenib metabolites also influence CYP isoenzymes having been shown capable of inhibiting CYP2C9 (the enzyme responsible for metabolizing warfarin), CYP2B6, CYP3A4 and CYP2C8 enzymes.[44]

Table 2 lists the agents potentially interacting with regorafenib and the effects of such interactions. In view of the potential pharmacological interactions, drug compatibility would have to be checked in all patients before starting regorafenib treatment.

Table 2 Major Drug Interactions with Regorafenib

Inducers of CYP3A4*	Inhibitors of CYP3A4^	CYP2C9 Inhibition#	UGT1A1 Inhibitor&
• Carbamazepine • Isoniazid • Phenobarbital • Phenytoin/ fosphenytoin • Rifampin • St. John's wort (Hypericum perforatum)	• Boceprevir • Clarithromycin • Conivaptan • Grapefruit juice • Ketoconazole • Indinavir • Itraconazole • Nefazodone • Nelfinavir • Posaconazole • Ritonavir • Saquinavir • Telaprevir • Telithromycin • Voriconazole	• Warfarin	• Irinotecan

Notes: *Inducers of CYP3A4 may decrease exposure to regorafenib and exposure to M-2 and M-5 metabolites may increase. ^Inhibitors of CYP3A4 may increase exposure to regorafenib and exposure to M-2 and M-5 metabolites may decrease. #Regorafenib inhibits CYP2C9; concomitant administration of drugs that are CYP2C9 substrates may result in increased exposure of that drug. &Regorafenib is a UGT1A1 inhibitor: concomitant use with irinotecan may result in increased irinotecan exposure.

Established Activity of Regorafenib in HCC

In preclinical studies, regorafenib was confirmed to be a strong inhibitor of Raf-1 and various receptor tyrosine kinases implicated in neovascularization and tumor progression, such as VEGFR-1-2-3, PDGFR, c-KIT, and FGFR. In xenograft models, this agent resulted in tumor growth inhibition of extracellular signal-regulated kinase phosphorylation, which could be revealed by an expressive decrease in microvessels density inside the tumor area.[31,45]

A Phase 2 study with 36 patients with HCC displayed both acceptable tolerability and proof of antitumor activity, with a median OS of 13.8 months and median TTP of 4.3 months.[46]

These results led to the conception of the RESORCE study, a Phase 3 placebo-controlled trial, which included patients who had progressed on sorafenib but were tolerating ≥400 mg/d for ≥20 of the last 28 days of treatment.[15] The last dose of sorafenib had to have been received

within the last 10 weeks prior to randomization, and 2 weeks washout from the last dose of sorafenib was required before starting regorafenib, and exclusion criteria included discontinuation of sorafenib due to toxicity. The trial was carried out in 152 centers across 21 different countries and four continents. Participants were assigned randomly (2:1) to 160 mg oral regorafenib or placebo once daily for 3 weeks followed by 1-week off. A total of 4 weeks constituted one full treatment cycle. All patients received the best supportive care.

The primary endpoint of the trial was overall survival time (time from randomization to death) and analyzed by intention to treat. The secondary endpoints were TTP, PFS, objective response rate (CR or PR), and disease control rate (CR, PR, or SD maintained for ≥6 weeks) as estimated by the investigators applying mRECIST and RECIST 1.1.

Patients were stratified by geographical region (Asia vs the rest of the world), the presence of macrovascular invasion (yes vs no), presence of extra-hepatic disease (yes vs no), α-fetoprotein concentration (<400 vs >400 ng/mL), and Eastern Cooperative Oncology Group performance status (0 vs 1). Following the screening of 843 patients, 573 were enrolled and randomized (379 to the regorafenib arm and 194 to the placebo arm).

A total of 216 patients were from Asia. The median treatment duration on sorafenib before commencing regorafenib was 7.8 months (IQR 4.2–14.5) in the regorafenib arm and 7.8 months (IQR: 4.4–14.7) in the placebo arm. The median time on regorafenib in this study was 3.6 months (IQR: 1.6–7.6) and 1.9 months (IQR: 1.4–3.9) on placebo. The mean daily regorafenib dose was 144.1 mg. With major relevance, a systemic therapy yielded a significantly higher OS in the second-line setting, with a 10.6 months median OS (95% CI: 9.1–12.1) on regorafenib compared with 7.8 months (6.3–8.8) for the placebo (HR: 0.63; 95% CI: 0.50–0.79; $p<0.0001$) with a 37% reduction in the risk of death. Median PFS was also significantly improved in the regorafenib group at 3.1 months (95% CI: 2.8–4.2) compared with 1.5 (1.4–1.6) months with the placebo group. This was a 54% reduction in the risk of progression or death (HR: 0.46; 95% CI: 0.37–0.56; $p < 0.0001$). The median TTP in the regorafenib arm was 3.2 months (2.9–4.2 95% CI) with regorafenib compared with 1.5 (1.4–1.6) months in the placebo arm (HR: 0.44; 95% CI: 0.36–0.55; $p<0.0001$). A total of 11% of patients treated with regorafenib compared with 4% in the placebo arm attained an objective response ($p=0.0047$). Two patients (1%) in the regorafenib arm versus zero patients in the placebo arm achieved a CR.

The rate of alpha-fetoprotein (AFP) response, defined as a ≥20% decrease in AFP from baseline at the start of cycle 3, was higher in patients treated with regorafenib than in those who received placebo.

The toxicity profile of regorafenib was similar to other TKIs, especially sorafenib. In the early phase studies, dose-limiting toxicities included bone marrow suppression and gastrointestinal toxicities.[31,42,44,45] In the subsequent Phase II trial of patients with HCC, 58% of patients experienced a grade 3 or higher adverse event (AE).[46] These included fatigue (17%), hand–foot skin reaction (14%), and diarrhea (6%). A total of 19% of patients stopped treatment due to AEs that were deemed treatment-related as per investigator opinion. In the following Phase III RESORCE study, AEs were reported in all patients treated with regorafenib (100%) and 179 of 193 patients were receiving placebo (93%).[15] The most frequent clinically relevant grade 3 or 4 treatment-emergent adverse events (TEAEs) were hypertension (15%), hand–foot skin reaction (HFSR) (13%), fatigue (9%), and diarrhea (3%). A total of 10% experienced a regorafenib-related serious AE and seven deaths (2%) were attributed to the study drug compared with 2% in the placebo group.

The only drug-related deaths due to liver failure were seen in the placebo group. A total of 6% of patients in the regorafenib treatment arm had grade 3 or higher TE bleeding events compared with 8% in the placebo arm.

A total of 255 (68%) of 374 patients in the regorafenib arm had dose interruptions or reductions due to AEs compared with 60 (31%) of 193 patients in the placebo arm.

Overall, regorafenib was well tolerated. Drug-related AEs led to dose interruptions or reductions in 202 (54%) patients in the regorafenib arm and 20 (10%) in the placebo arm, discontinuation due to a treatment-related AEs was relatively low at 39 (10%) in the regorafenib arm compared with 7 (4%) in the placebo arm.

The most common AEs leading to treatment discontinuation more frequently seen with regorafenib were i) increase in the liver enzyme aspartate transaminase (AST) (8 [2%] of 374 patients vs 3 [2%] in the placebo group), ii) hand–foot skin reaction (7 [2%] vs none), and iii) an increase in alanine transaminase (ALT) (4 [1%] vs none).

Further subanalyses of the RESORCE study have been performed and showed that I) a longer survival follow-up almost 1 year after the primary analysis confirmed the primary OS results;[47] II) by comparing tumor response

and progression in the RESORCE trial by mRECIST and RECIST 1.1, although a slightly higher response rate was observed using mRECIST criteria, rates of PFS, TTP, and disease control were not different when assessed by investigators using mRECIST or RECIST 1.1;[48,49] III) in an exploratory analysis, aimed at validating concept of progression profile in a global cohort of previously sorafenib treated patients and assessing the impact of regorafenib on survival by looking at previous progression, regorafenib provides an OS benefit regardless of progression pattern;[50] IV) in a post-hoc exploratory analysis, patients developing HFSR under regorafenib tended to have improved OS (Median OS, months 14.1 months [95% CI 11.7, 16.5] versus 6.6 [5.0, 8.5]), as was previously shown for sorafenib.[51,52]

Finally, an exploratory analysis of the RESORCE study reported that patients who received the sorafenib-regorafenib sequence achieved a never-before-demonstrated outcome, with a median OS of 26 months.[53]

Also, the analysis showed that regorafenib treatment resulted in a clinical benefit regardless of the sorafenib last dose or TTP on sorafenib.

Real-Life Clinical Practice of Regorafenib Treatment in HCC

Since the approval of regorafenib in 2017, clinical practice studies have reported results, which are however still limited, on the safety and efficacy profile of regorafenib in real-life experience.[54–57]

REFINE (NCT03289273) is an ongoing observational study that recruited patients with HCC for whom a decision to treat with regorafenib was made by the treating physician before enrollment, according to the local health authority approved label.[54]

The findings from the interim analysis carried out after the first 500 enrolled patients were presented during the 2020 International Liver Cancer Association (ILCA).[55]

REFINE had a broader patient population compared with RESORCE, which reflects the less stringent inclusion criteria of the real-world study. Most patients (67%) had Child–Pugh class A liver function; 11% and 1% had Child–Pugh class B and C liver function, respectively (the Child–Pugh score was missing or not evaluable in 21% of patients).

The proportions of patients with Eastern Cooperative Oncology Group performance status (ECOG PS) 0, 1, and 2–4 were 42%, 40%, and 5%, respectively (the ECOG PS was missing or not evaluable in 13% of patients). Most patients (98%; n=490) had received prior systemic therapy; 97% (n=482) had received prior sorafenib. Regorafenib was second-line treatment in 81% of patients (n=403), third-line or higher in 17% (n=87), and first-line in 2% (n=8).

Of the 403 patients who received regorafenib second-line, 398 (99%) had received prior sorafenib. Among all treated patients (N=498), 57% (n=286) initiated regorafenib at a daily dose of 160 mg, 13% (n=63) at 120 mg, 28% (n=141) at 80 mg, and 2% (n=8) at 40 mg.

In the 482 patients who received sorafenib in any prior line of therapy, the median duration of prior sorafenib was 4.8 months (interquartile range 2.5–9.6), 45% of patients (n=216) had a last daily sorafenib dose of 800 mg, 8% of patients (n=40) had an AE leading to sorafenib discontinuation (defined as sorafenib-intolerant patients) and, at study entry, the proportions of patients with Child–Pugh class A, B, and C liver disease were 67%, 12%, and 1%, respectively. Regarding safety, among all regorafenib treated patients (n=498), the most frequent TEAEs (any grade) were HFSR (30%), diarrhea (21%), fatigue (16%), and decreased appetite (14%) while in sorafenib intolerant patients, the most frequent TEAEs (any grade) with regorafenib were diarrhea, HFSR, abdominal pain, and decreased appetite.

Investigators also evaluated the OS by Child–Pugh class and ALBI grade at study entry in the patients who had received sorafenib previously. The median OS was 16.0 months among the Child–Pugh class A group (95% CI, 13.1–18.8) versus 8.0 months among the Child–Pugh class B group (95% CI, 5.2-not evaluable [NE]). The median OS among those with ALBI grades 1, 2, and 3, respectively, was 19.6 months (95% CI, 14.8–19.6), 10.5 (95% CI, 8.7–16.0), and 3.1 months (95% CI, 1.6–8.7).

Regorafenib confirmed survival benefit regardless of the rate of disease progression during the preceding treatment with sorafenib or since the last sorafenib dose in a retrospective study of safety and efficacy in Korean patients where data were consistent with those from the RESORCE trial.[56]

In a subsequent multicenter, retrospective analysis of 440 patients who received prior sorafenib and were treated with regorafenib as the second (69.3%), third (26.1%), and fourth to seventh (4.5%) lines of therapy in nine tertiary referral hospitals in Korea, real-life clinical outcomes were consistent with the RESORCE trial results and regorafenib

related HFSR resulted to be significantly associated with better OS.[57]

Interestingly, ICIs were given in 115 patients (26.1%) prior to regorafenib and there were no differences in PFS and OS with regorafenib according to the prior use of ICIs.

A clinically relevant aspect emerging from some clinical practice studies is the importance of the patient's physical status and residual liver function after first-line failure. These parameters affect the rate of patients eligible for switching to second-line agents after radiologic progression with first-line sorafenib treatment.

A Canadian study characterized subsequent therapies received by HCC patients following sorafenib and determined the rate of patients eligible for novel therapies if strict eligibility criteria (SEC) (as defined in their respective trials) were used, compared to more liberal modified eligibility criteria (MEC, including Child–Pugh B7 and ECOG 2).[58]

A total of 730 patients were identified with 172 (23.6%) receiving subsequent treatment (regorafenib, cabozantinib or ramucirumab). Patients receiving subsequent treatment had longer overall survival than patients who did not (12.1 versus 3.3 months; $p<0.001$). Using SEC, only 13.1% of patients would be eligible for second-line treatment. Extending accessibility to patients meeting the MEC increased the eligibility rate to 31.7%.

The highest ineligibility for regorafenib was determined by study-specific criteria, including intolerance to sorafenib (28%).

Thus, the study showed that only a limited proportion of HCC real-world patients would be eligible for cabozantinib, regorafenib, or ramucirumab if SEC in clinical trials were followed, while more than double would be eligible if MEC were followed. Patients who received subsequent treatment had improved OS, irrespective of whether they encountered SEC or MEC.

A small Japanese retrospective study reported that in clinical practice only about 30% of patients refractory to first-line sorafenib therapy are eligible for second-line regorafenib treatment.[59] The main reasons that patients could not be treated with regorafenib were their intolerance to sorafenib and deterioration of liver function.

This and other real-life studies highlight that to prolong the prognosis with the use of effective second-line agents, it is important to maintain liver function before and during both previous transarterial and first-line therapies.[60]

Adequate liver function reserve and ECOG performance status during treatment with sorafenib accounted for the efficacy and improved outcome of the following treatment.[60–62]

This is substantiated by the finding that the novel biomarker of liver reserve function, albumin-bilirubin (ALBI) grade, was able to successfully detect regorafenib candidates and that a median OS of 15.6 months was obtained in the selected cohort compared to 6.8 months for non-candidates.[63,64]

Takada et al confirmed in a recent study that a more accurate estimation of liver function has emerged as an essential requirement in this setting.[65] They showed that at the moment of failure of first-line sorafenib, the criteria for inclusion in the RESORCE study were not just the baseline ALBI score (−2.33; OR 2.5, p=0.01) but also the level of variation in liver function after four weeks of treatment with sorafenib (<0.255; OR 4.9, $p < 0.001$).

Besides, Yukimoto et al showed that an ALBI score of −2.53 at the moment of sorafenib initiation was useful as a threshold value for the prediction of regorafenib eligibility following the failure of sorafenib.[64]

Similarly, Moriguchi et al showed that ALBI grade at the initiation of sorafenib therapy is a significant factor that contributed to the maintenance of Child–Pugh grade A and ECOG-PS ≤1 upon sorafenib discontinuation and is a good indicator of the possibility of the introduction of second-line therapy after sorafenib for HCC.[66]

Accordingly, a recent small retrospective study suggests that regorafenib's clinical outcomes and increased frequency of severe adverse events would discourage its use in Child–Pugh B patients with ALBI grade 3.[67]

Since there are still no proven biomarkers in clinical practice to guide systemic therapy, a Japanese study aimed to evaluate relative dose intensity (RDI, defined as the ratio of administered dose to planned dose) and the association between RDI and OS in patients with unresectable HCC.[68] Patients with first-month RDI ≥ 50% were shown to have significantly better OS and PFS than those with first-month RDI < 50% (HR 0.19 [CI 0.08–0.48], p=0.0004 and HR 0.2 [CI 0.08–0.52] p=0.0008), and a first month-RDI ≥ 50% (HR 0.18 [CI 0.06–0.55] p=0.002) and a hand-foot skin reaction (HR 0.03 [CI 0.008–0.16] $p < 0.0001$) were independently correlated with OS.

Therefore, Sorafenib-regorafenib sequential treatment is effective and well-tolerated in Japanese patients with unresectable hepatocellular carcinoma. A first month-RDI of ≥50% regorafenib has proven clinical relevance and, if confirmed in larger studies, could be a useful tool to assist second-line therapy.

Finally, regorafenib also proved to be effective in sorafenib-tolerant patients with recurrent HCC after liver transplantation who develop progression in is a retrospective, multicenter, international study with a median OS of 12.9 months after starting regorafenib and 38.4 months after initiation of sorafenib (18.5–58.4 95% CI) for sorafenib-regorafenib sequential treatment.

The AEs reported in the study were not only similar to those reported in the registration study but also comparable to those reported in similar patients receiving previous sorafenib treatment.[69]

Prognostic Markers Associated with Response to Regorafenib

Following the development of new effective systemic therapies for HCC, the current challenge is the correct selection of patients to orient the appropriate choice of treatment.

The identification of relevant predictive markers for clinical outcomes associated with regorafenib treatment is critical. However, to date, no established biomarkers have been identified.

In the absence of clinical/biological predictors to identify potentially responsive patients, a retrospective biomarker analysis was performed on patients enrolled in the RESORCE trial to identify biomarkers potentially predictive of benefit for regorafenib in HCC.[70]

Plasma and tumor samples from RESORCE study participants were evaluated in 567 patients (374 regorafenib arm and 193 placebo arm) to identify genetic, microRNA (miRNA), and protein biomarkers associated with response to regorafenib.

Remarkably, nine miRNAs (MIR30A, MIR122, MIR125B, MIR200A, MIR374B, MIR15B, MIR107, MIR320, and MIR645) plasma levels were identified as significantly related to overall survival time with regorafenib. Also, five proteins were identified as predictors of the benefit of regorafenib treatment for OS (angiopoietin 1 [ANG-1], cystatin B, transforming growth factor-beta 1 latency-associated peptide [LAP TGF-b1], oxidized low-density lipoprotein receptor 1 [LOX-1], C-C motif chemokine ligand 3 [MIP-1a] with decreased levels associated with the benefit of regorafenib treatment).

Currently, this is the only study that provides a possible biomarker-guided strategy for the identification of patients potentially responsive to regorafenib, but it still needs validation in further studies.

It has been suggested that Tie2 is a potential circulating biomarker of tumor vascular response for VEGF inhibitors assuming that Tie2 originates from the tumor blood vessels.[71]

Since oncological use of anti-angiogenic VEGF inhibitors has been limited by the lack of informative biomarkers, circulating Tie2 could be a candidate tumor vascular response biomarker for VEGF inhibitors.

Interestingly, during regorafenib treatment a dynamic modification of plasma angiogenic components has been reported: low baseline levels of angiopoietin-2 and Tie-2 appear to be related to a better prognosis and early modulation of Ang-2 levels may be predictive of response to regorafenib in patients with metastatic colorectal cancer.

Such results would justify an exploratory study to confirm this prognostic correlation in HCC patients.[72]

Combination of Regorafenib with Other Systemic Agents

In recent years, encouraging data that would promote the combination of antiangiogenic effects of regorafenib with ICIs to optimize and increase the response rate of the two individual therapies have been reported.[73,74]

Since TME, a key determinant of tumor growth and metastasis is characterized by multiple counterparts including immune and non-immune cell populations as well as non-cellular components, combination use of current TKIs with immunotherapy has been investigated to maximally exploit the therapeutic benefit.[75,76]

Regorafenib within the sub-micromolar range was found to induce M1 macrophage polarization and enhance CD8+ T cells proliferation and activation of (Figure 2). Besides, in vivo studies using regorafenib at low-dose (3–5 mg/kg/day), representing approximately 50% of the recommended single-agent dosage in the clinic showed synergistic antitumor efficacy with anti-PD-1 therapy.[22]

Identifying the optimal immunomodulatory-effects of targeted-agents is therefore crucial for the development of combination immunotherapy to enhance the therapeutic index and to tailor the use of targeted drugs to their biologically active and clinically significant dosage.[21]

The limitations of monotherapy approaches in HCC have led to the development of combination strategies using anti-VEGFR and anti-PD1/PD-L1 to address mechanisms of treatment resistance and achieve synergy by increasing tumor infiltration of effector T cells.[22]

One of the key elements of the efficacy of combination therapies with anti-VEGF, such as regorafenib plus immunotherapy, is to maintain a functional (normalized) vasculature and reduce hypoxia. Of note, recent studies have also shown that antitumor immune responses and vascular normalization can be reciprocally regulated by CD4+ T effector cells in other cancers.[40,77–81]

In HCC, dual VEGFR-2/PD1 blockade using antibodies has recently been shown to normalize tumor vasculature and induce antitumor immunity in a preclinical animal model with underlying liver damage.[82]

The use of combination therapy with anti-VEGF (bevacizumab) and anti-PD-L1 (atezolizumab) monoclonal antibodies has been shown to significantly increase survival compared with sorafenib in the randomized phase III IMBRAVE150 study.[14]

Therefore, combination approaches using multitarget TKI, such as regorafenib, that are not restricted to VEGF alone, could achieve higher efficacy outcomes and are currently under investigation.

In a preclinical study, it has been reported that in a combination treatment with anti-PD-1 antibody, regorafenib can significantly enhance PD1 blockade effects in a dose-dependent manner in HCC models.[40]

The benefit was due to the activity of the two agents on both normalizations of the HCC vasculature and stimulation of anti-tumor immunity. The combination treatment inhibited STAT3 activity and increased the expression of the chemokine CXCL10, which increased both tumor penetration and survival of activated CD8 T cells.

This concept is clinically relevant for the future design of combination treatment strategies in HCC patients.

The potential synergistic antitumor efficacy of regorafenib with anti-PD1 therapy has been also shown in a study of an orthotopic HCC model demonstrating that regorafenib may modulate macrophage polarization, increase T cell activation, and thereby enhance the efficacy of anti-PD1 therapy for HCC.[83]

In a recent open-label, dose-escalation Ib study another TKIs/ICI combination treatment based on regorafenib plus pembrolizumab, an anti-PD-1 monoclonal antibody, was tested in patients with advanced HCC who received no previous systemic treatment (NCT03347292).[84]

In the first cohort, patients underwent regorafenib 120 mg/day PO for three weeks on/1 week off with pembrolizumab 200 mg IV q 3 weeks. Thereafter, the regorafenib dose could be increased (160 mg) or lowered (80 mg) according to the modified toxicity probability interval design, while the dosage of pembrolizumab was steady. The primary endpoints were tolerability and safety. Secondary aims were to define the maximum tolerated dose (MTD) and recommended phase 2 dose and to assess anti-tumor efficacy. Twenty-nine patients received regorafenib 120 mg dosage. The median age was 65 years (range 32–81), 41% and 55% of patients had BCLC stage B and C respectively, while 100% were Child–Pugh A class; ECOG status 1/0 was 28%/72%. Dose-limiting toxicities were reported in 4/18 evaluable patients: grade 3 raised AST/ALT with grade 2 elevated bilirubin (n=2); grade 3 rash (n=2). The MTD of regorafenib in the combined treatment was 120 mg.

There were no grade 5 TEAEs. Dose modifications (interruption and/or dose reduction) of regorafenib/pembrolizumab for drug-related TEAEs were reported in 59%/31% of patients.

Of 23 assessable patients, 7 (30%) exhibited a partial response and 14 (61%) showed stable disease (according to RECIST v1.1). One additional patient had a partial response (according to mRECIST). Thus, the combined treatment with regorafenib plus pembrolizumab as first-line therapy of advanced HCC showed encouraging signs of anti-tumor activity and safety profile.

Enrolment has been continued and is ongoing at regorafenib 120 mg dose.

Nivolumab is a human immunoglobulin G4 (IgG4) monoclonal antibody to the programmed death (PD)-1 receptor, which blocks the interaction with PD-ligand (PD-L)1/PD-L2 thus resuming T-cell-mediated antitumor effects, which has been approved in 2017 for the second-line treatment of HCC patients who have been previously treated with sorafenib.[18]

A phase I/IIa trial (NCT04170556) is ongoing and is aimed to evaluate the effects of nivolumab and regorafenib but considering the potential impact of the interaction of drugs and enhanced severity and/or frequency of AEs.[85]

Thus, regorafenib will be administered as monotherapy during the first 2 cycles (each cycle is 3 weeks on plus 1 week off) of treatment to enhance T cell trafficking and infiltration into the tumor bed to increase the benefits of anti-PD-PD-L1.

Tislelizumab, is a humanized monoclonal antibody directed against PD-1, currently tested for hematological cancers and advanced solid tumors.[86]

An ongoing phase 2 study (NCT04183088) will investigate the efficacy and safety of the combined tislelizumab with regorafenib as first-line treatment for advanced HCC.[87]

This trial consists of 2 parts. Part 1 consists of a single-arm study and eligible patients will be assigned

tislelizumab 200 mg intravenously on day 1 every 3 weeks plus regorafenib (80 mg/d). Part 2 is a randomized study. Subjects will be 1:1 randomized to two treatment arms: (1) tislelizumab and regorafenib combined treatment as used in part 1, versus (2) regorafenib and placebo. For patients in group 2, when imaging assessment shows SD or PD, according to RECIST v1.1 criteria, the therapeutic strategy will be shifted to tislelizumab + regorafenib combination schedule.

Lastly, a multicenter, open-labeled prospective phase Ib trial (NCT03475953) investigating three dosage levels of Regorafenib combined with Avelumab, a human IgG1 monoclonal antibody that targets PD-L1, in both advanced and metastatic solid tumors (including HCC), is currently being recruiting.[88]

Ongoing clinical trials based on combined regorafenib/ICI agents are reported in Table 3.

In addition to immunotherapy, other combination treatments of regorafenib with agents acting on parallel and complementary pathogenic pathways have also been reported in preclinical cancer models.[22]

Annexin A3 (ANXA3) is well known to have a key role in enhancing tumor aggressiveness, preventing apoptosis, and promoting prosurvival autophagy in sorafenib-resistant HCC cells.

Tong et al demonstrated in in-vivo models of sorafenib unresponsive HCC that co-administration of regorafenib and an anti-ANXA-3 monoclonal antibody can potentiate apoptotic induction by abrogating autophagy.[89] Likewise, navitoclax, a specific inhibitor of the anti-apoptotic proteins Bcl-2 and Bcl-xL, enhanced the regorafenib sensitivity of Hep3B and HepG2 cells, as evidenced by enhanced apoptotic features.[90]

The potential benefit of regorafenib has also been tested in combination treatment with TACE.

Regorafenib-loaded Poly(lactide-co-glycolide) (PLGA) microspheres for improvement of transarterial chemoembolization (TACE) therapeutic effects, which can sustainably deliver regorafenib to limit proangiogenic responses in liver tumors after TACE, has been recently developed. The fabricated regorafenib microspheres provided sustained drug release for more than 30 d in vitro and in vivo after TACE. The study demonstrated that the new regorafenib microspheres, as local drug delivery combined with TACE, may enhance the therapeutic potency of TACE for the treatment of HCC and has promising clinical implications in the future.[91]

Discussion

Recent studies have begun to decode the complexity of the HCC immune microenvironment, such as the function and subsets of different immune cells in the liver, including T and B cells, macrophages, neutrophils, NK cells, dendritic cells, myeloid-derived suppressor cells, cancer-associated fibroblasts, and the active interplay between immune cells and the cancer ecosystem in promoting angiogenesis.[92]

This evidence constitutes a strong rationale supporting a therapeutic strategy that simultaneously targets the main pathogenic pathways that sustain tumor proliferation, spread, and neoangiogenesis as well as the immune mechanisms that allow tumor cells to evade immune suppression.[41]

This combined approach is highly likely to lead to an enhancement of anti-tumor therapies with maximized response rates and the possibility of achieving not only disease stabilization but also tumor mass shrinkage with a higher objective response rate.

The combined use of atezolizumab with bevacizumab in first-line unresectable HCC has newly demonstrated a superior benefit over sorafenib in a recently published phase 3 trial, thus confirming that by targeting simultaneously the pathogenic pathways that support tumor growth, spread and neoangiogenesis, on the one hand, and immunosuppression and tumor-induced immune evasion on the other, the efficacy of anti-tumor treatments can be significantly improved.[14]

It would also be of potential interest to test whether a treatment strategy including another ICI (anti-CTLA-4) or a broad-spectrum multikinase inhibitor added to a PD-1 or PD-L1 inhibitors, could provide additional benefits. This strategy has been successfully studied in more detail in other malignant diseases such as renal cell carcinoma.[93–96]

Specifically, focused studies are indispensable as feasibility and comparison of these studies with HCC patients is not practicable due to diversity in tumor biology and underlying liver disease characterizing HCC patients.

Another clinically relevant question will be to assess the suitability of such therapies for patients with Child–Pugh class B (especially B7), which is currently still limited because these patients have been ruled out or only accounted for less than 10% of patients in studies.

However, these therapies are utilized in clinical practice with not as strict inclusion criteria as the respective phase 3

Table 3 Ongoing Clinical Trials with Regorafenib-Based Combination Treatments (www.clinicaltrials.gov)

Clinical Trials Identifier	Official Title	Phase	Therapy Line	Intervention/Treatment	Status
NCT04183088	Regorafenib Plus Tislelizumab as First-line Systemic Therapy for Patients With Advanced Hepatocellular Carcinoma	II	First	• Tislelizumab+regorafenib for part 1. • Tislelizumab+regorafenib for group 1 of part 2. • Placebo+regorafenib for group 2 of part 2.	Not yet recruiting
NCT04170556	The GOING Study: Regorafenib Followed by Nivolumab in Patients With Hepatocellular Carcinoma Progressing Under Sorafenib	I/IIa	Second^	• Regorafenib 160 mg/day 3 weeks on and 1 week off. • Nivolumab at the dose of 1.5 mg/kg, 3 mg/kg or 240 mg/infusion every 2 weeks. Dose will be adjusted depending on the incidence of adverse events.	Recruiting
NCT04310709	Phase II Study of Regorafenib-nivolumab Combination Therapy for Chemotherapy-naïve Patients With Unresectable or Metastatic Hepatocellular (RENOBATE)	II	First	• Nivolumab 480 mg IV on Day 1, every 4 weeks. • Regorafenib 80 mg per oral once daily for 21 consecutive days starting on Day 1, every 4 weeks.	Recruiting
NCT04777851	Phase III, Multicenter, Randomized, Open-Label Trial to Evaluate Efficacy and Safety of Regorafenib in Combination With Nivolumab Versus TACE for First-Line Treatment of Intermediate-Stage HCC With Beyond Up-to-7 Criteria	III	First	**Investigational arm:** regorafenib at a dose of 90 mg orally once per day (on days 1 to 21 of a 28-day cycle), in combination with nivolumab 480 mg using 30-minutes intravenous infusion (on day 1 of a 28-day cycle, every 4 weeks). **Control arm:** Patients will be treated with transarterial chemoembolization (TACE) "on-demand" according to the clinical site's standards, with the goal of controlling all known liver lesions. Either conventional TACE (cTACE) or drug-eluting bead transarterial chemoembolization (DEB-TACE) may be used (as long as it is consistently applied for all patients at a given clinical site).	Not yet recruiting
NCT04718909	Regorafenib Combined With Sintilimab Versus Regorafenib Alone as the Second-line Treatment for Unresectable Hepatocellular Carcinoma	II	Second	• **Experimental Arm:** Regorafenib: 160 mg p.o. qd for 3 weeks of every 4 week cycle (ie, 3 weeks on, 1 week off). Sintilimab: 200mg i.v. q3w. • **Active Comparator:** Regorafenib: 160 mg p.o. qd for 3 weeks of every 4 week cycle (ie, 3 weeks on, 1 week off).	Recruiting

(*Continued*)

Table 3 (Continued).

Clinical Trials Identifier	Official Title	Phase	Therapy Line	Intervention/Treatment	Status
NCT03475953	A Phase I/II Study of Regorafenib Plus Avelumab in Solid Tumors	I/II	≥ 1 previous line (s) of systemic therapy	• 3 dose levels of Regorafenib given in combination with Avelumab followed by 7 phase II trials to evaluate the association of Regorafenib at the RP2D[§] with Avelumab in 7 distinct settings (advanced or metastatic tumors).	Recruiting
NCT04696055	An Open-Label Study of Regorafenib in Combination With Pembrolizumab in Patients With Advanced or Metastatic Hepatocellular Carcinoma (HCC) After PD1/PD-L1 Immune Checkpoint Inhibitors	II	Prior 1L immunotherapy with a PD-1/PD-L1 checkpoint inhibitor administered either as monotherapy or in combination with other therapies	• Pembrolizumab 400 mg to be administered as an intravenous (IV) infusion every 6 weeks (Q6W). • Regorafenib will be given orally (p.o.) at a starting dose of 90 mg QD for 3 weeks of every 4 weeks (ie, 3 weeks on, 1 week off). If the starting dose of 90 mg daily is well tolerated the dose should be escalated to 120 mg starting after the first 4-week cycle of regorafenib.	Recruiting
NCT03347292	A Multicenter, Non-randomized, Open-label Dose Escalation Phase Ib Study of Regorafenib in Combination With Pembrolizumab in Patients With Advanced Hepatocellular Carcinoma (HCC) With no Prior Systemic Therapy	I	First	• Dose escalation: The regorafenib starting dose will be 120 mg q.d. (once daily) 3 weeks on/1 week off in combination with the recommended dose of pembrolizumab (200 mg Q3W). Pembrolizumab dose will not be escalated or de-escalated. • Dose expansion: Dose expansion cohorts will continue to be expanded until the sample size of 30–35 patients per cohort is reached.	Active, not recruiting

Note: ^Patients progressing under first-line sorafenib.
Abbreviation: [§]RP2D, recommended phase II dose.

studies.[21,60,67] For instance, in the prospective observational study (REFINE) of regorafenib in HCC patients, 11% of treated patients had Child–Pugh B liver function and 28% of total patients were initiated on 80 mg of regorafenib rather than the standard 160 mg dose.[21] These dose modifications to attenuate TKI-related AEs without affecting efficacy have been prospectively analyzed in metastatic colorectal cancer, where a dose-escalation approach to regorafenib showed favorable AEs and comparable therapeutic efficacy to the entire dose.[97] Equally, sorafenib at 200 mg proved more tolerable than the 400 mg dose with comparable efficacy in a large retrospective study.[98,99]

Regorafenib, due to its broad spectrum of inhibition of tumor angiogenesis and its favorable effects on macrophage polarization and cytotoxic CD8+ lymphocyte activity, is a strong candidate for combination therapy strategies with ICIs.A recent meta-analysis based mainly on real-world studies investigating regorafenib assecond-line therapy after sorafenib failure confirms the promising favorable outcomes observed with the RESORCE trial and demonstrates that regorafenib provides both valid and safe treatment strategy in patients with intermediate/advanced HCC who exhibit disease progression on sorafenib.[100]

In the next future, new clinical trials for HCC patients should be aimed at investigating the potential benefit and synergistic effects of regorafenib with ICIs.[60,]

A key step in the development of optimal systemic treatment strategies for HCC remains the identification of clinical-biological markers of efficacy and thus predictive biomarker's discovery is critical for the fine-tuning of regorafenib treatment.[70]

New combined therapeutic approaches of regorafenib with other ICIs or TKIs provide an interesting opportunity for continued research and in the near future is expected to be a breakthrough for patients with unresectable HCC. It will therefore be critical to have in-depth knowledge of the pharmacological characteristics of each drug and the most appropriate management of possible AEs to achieve maximum therapeutic benefit.

Funding

The authors received no financial support to produce this manuscript.

Disclosure

Fabio Piscaglia has received personal fees for speakers' bureaus and advisory boards and for being a consultant from Alkermes, AstraZeneca, Bayer, Bracco, Bristol Myers Squibb, Eisai, GE Healthcare, Ipsen, La Force Guerbet, Roche and Tiziana Life Sciences.

Francesco Tovoli reports personal fees from Bayer AG, Guerbet, and Ipsen, outside the submitted work; consultant for Bayer, speaker bureau honoraria from MSD, and grant from Ipsen.

The authors reported no other potential conflicts of interest for this work.

References

1. Global Cancer Observatory (GCO)@GLOBOCAN_GCO. Available from: https://gco.iarc.fr/today/home. Accessed April 20, 2021.
2. El-Serag HB. Epidemiology of viral hepatitis and hepatocellular carcinoma. *Gastroenterology*. 2012;142(6):1264–1273. doi:10.1053/j.gastro.2011.12.061
3. White DL, Thrift AP, Kanwal F, Davila J, El-Serag HB. Incidence of hepatocellular carcinoma in all 50 United States, from 2000 through 2012. *Gastroenterology*. 2017;152(4):812–820, e5. doi:10.1053/j.gastro.2016.11.020
4. Singal AG, Lampertico P, Nahon P. Epidemiology and surveillance for hepatocellular carcinoma: new trends. *J Hepatol*. 2020;72(2):250–261. doi:10.1016/j.jhep.2019.08.025
5. Siegel AB, Zhu AX. Metabolic syndrome and hepatocellular carcinoma: two growing epidemics with a potential link. *Cancer*. 2009;115:5651–5661. doi:10.1002/cncr.24687
6. Baffy G, Brunt EM, Caldwell SH. Hepatocellular carcinoma in non-alcoholic fatty liver disease: an emerging menace. *J Hepatol*. 2012;56:1384–1391. doi:10.1016/j.jhep.2011.10.027
7. Dyson J, Jaques B, Chattopadyhay D, et al. Hepatocellular cancer: the impact of obesity, type 2 diabetes and a multidisciplinary team. *J Hepatol*. 2014;60:110–117. doi:10.1016/j.jhep.2013.08.011
8. Singal AK, Singh A, Jaganmohan S, et al. Antiviral therapy reduces risk of hepatocellular carcinoma in patients with hepatitis C virus-related cirrhosis. *Clin Gastroenterol Hepatol*. 2010;8:192–199. doi:10.1016/j.cgh.2009.10.026
9. Morgan TR, Ghany MG, Kim HY, et al. Outcome of sustained virological responders with histologically advanced chronic hepatitis C. *Hepatology*. 2010;52:833–844. doi:10.1002/hep.23744
10. Santi V, Buccione D, Di Micoli A, et al. The changing scenario of hepatocellular carcinoma over the last two decades in Italy. *J Hepatol*. 2012;56(2):397–405. doi:10.1016/j.jhep.2011.05.026
11. Simmons B, Saleem J, Heath K, Cooke GS, Hill A. Long-term treatment outcomes of patients infected with hepatitis C virus: a systematic review and meta-analysis of the survival benefit of achieving a sustained virological response. *Clin Infect Dis*. 2015;61:730–740. doi:10.1093/cid/civ396
12. Llovet JM, Ricci S, Mazzaferro V, et al. Sorafenib in advanced hepatocellular carcinoma. *N Engl J Med*. 2008;359:378–390. doi:10.1056/NEJMoa0708857
13. Kudo M, Finn RS, Qin S, et al. Lenvatinib versus sorafenib in first-line treatment of patients with unresectable hepatocellular carcinoma: a randomised phase 3 non-inferiority trial. *Lancet*. 2018;391:1163–1173. doi:10.1016/S0140-6736(18)30207-1
14. Finn RS, Qin S, Ikeda M, et al. Atezolizumab plus Bevacizumab in Unresectable Hepatocellular Carcinoma. *N Engl J Med*. 2020;382(20):1894–1905. doi:10.1056/NEJMoa1915745
15. Bruix J, Qin S, Merle P, et al. Regorafenib for patients with hepatocellular carcinoma who progressed on sorafenib treatment (RESORCE): a randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet*. 2017;389:56–66. doi:10.1016/S0140-6736(16)32453-9
16. Abou-Alfa GK, Meyer T, Cheng AL, et al. Cabozantinib in patients with advanced and progressing hepatocellular carcinoma. *N Engl J Med*. 2018;379:54–63. doi:10.1056/NEJMoa1717002
17. Zhu AX, Kang YK, Yen CJ, et al. Ramucirumab after sorafenib in patients with advanced hepatocellular carcinoma and increased alpha-fetoprotein concentrations (REACH-2): a randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet Oncol*. 2019;20:282–296. doi:10.1016/S1470-2045(18)30937-9
18. El-Khoueiry AB, Sangro B, Yau T, et al. Nivolumab in patients with advanced hepatocellular carcinoma (CheckMate 040): an open-label, noncomparative, Phase 1/2 dose escalation and expansion trial. *Lancet*. 2017;389:2492–2502. doi:10.1016/S0140-6736(17)31046-2
19. Zhu AX, Finn RS, Edeline J, et al. Pembrolizumab in patients with advanced hepatocellular carcinoma previously treated with sorafenib (KEYNOTE-224): a non-randomised, open-label phase 2 trial. *Lancet Oncol*. 2018;19(7):940–952. doi:10.1016/S1470-2045(18)30351-6
20. Yau T, Kang Y, Kim T, et al. Efficacy and safety of nivolumab plus ipilimumab in patients with advanced hepatocellular carcinoma previously treated with sorafenib: the CheckMate 040 Randomized Clinical Trial. *JAMA Oncol*. 2020;6(11):e204564. doi:10.1001/jamaoncol.2020.4564
21. Lim HY, Merle P, Finn RS, et al. Regorafenib in patients with unresectable hepatocellular carcinoma (uHCC) in routine clinical practice: interim analysis of the prospective, observational REFINE trial. *J Clin Oncol*. 2020;38(suppl 4):542. doi:10.1200/JCO.2020.38.4_suppl.542
22. Cheng AL, Hsu C, Chan SL, Choo SP, Kudo M. Challenges of combination therapy with immune checkpoint inhibitors for hepatocellular carcinoma. *J Hepatol*. 2020;72:307–319. doi:10.1016/j.jhep.2019.09.025

23. Fondevila F, Méndez-Blanco C, Fernández-Palanca P, González-Gallego J, Mauriz JL. Anti-tumoral activity of single and combined regorafenib treatments in preclinical models of liver and gastrointestinal cancers. *Exp Mol Med*. 2019;51:1–15. doi:10.1038/s12276-019-0308-1
24. Wilhelm SM, Dumas J, Adnane L, et al. Regorafenib (BAY 73–4506): a new oral multikinase inhibitor of angiogenic, stromal and oncogenic receptor tyrosine kinases with potent preclinical antitumor activity. *Int J Cancer*. 2011;129:245–255. doi:10.1002/ijc.25864
25. Wai Ling Khoo TS, Rehman A, Olynyk JK. Tyrosine kinase inhibitors in the treatment of hepatocellular carcinoma. In: Tirnitz-Parker JEE, editor. *Hepatocellular Carcinoma [Internet]*. Brisbane (AU): Codon Publications; October 24, 2019. Chapter 7.
26. Eso Y, Marusawa H. Novel approaches for molecular targeted therapy against hepatocellular carcinoma. *Hepatol Res*. 2018;48 (8):597–607. doi:10.1111/hepr.13181
27. Kudo M. Targeted and immune therapies for hepatocellular carcinoma: predictions for 2019 and beyond. *World J Gastroenterol*. 2019;25:789–807. doi:10.3748/wjg.v25.i7.789
28. Mody K, Abou-Alfa GK. Systemic therapy for advanced hepatocellular carcinoma in an evolving landscape. *Curr Treat Options Oncol*. 2019;20:3. doi:10.1007/s11864-019-0601-1
29. Abou-Elkacem L, Arns S, Brix G, et al. Regorafenib inhibits growth, angiogenesis, and metastasis in a highly aggressive, orthotopic colon cancer model. *Mol Cancer Ther*. 2013;12 (7):1322–1331. doi:10.1158/1535-7163.MCT-12-1162
30. Schmieder R, Hoffmann J, Becker M, et al. Regorafenib (BAY 73–4506): antitumor and antimetastatic activities in preclinical models of colorectal cancer. *Int J Cancer*. 2014;135:1487–1496. doi:10.1002/ijc.28669
31. Zopf D, Fichtner I, Bhargava A, et al. Pharmacologic activity and pharmacokinetics of metabolites of regorafenib in preclinical models. *Cancer Med*. 2016;5:3176. doi:10.1002/cam4.883
32. Arai H, Battaglin F, Wang J, et al. Molecular insight of regorafenib treatment for colorectal cancer. *Cancer Treat Rev*. 2019;81:101912. doi:10.1016/j.ctrv.2019.101912
33. Hu W, Li X, Zhang C, Yang Y, Jiang J, Wu C. Tumor-associated macrophages in cancers. *Clin Transl Oncol*. 2016;18:251–258. doi:10.1007/s12094-015-1373-0
34. Kang FB, Wang L, Li D, Zhang YG, Sun DX. Hepatocellular carcinomas promote tumor-associated macrophage M2-polarization via increased B7-H3 expression. *Oncol Rep*. 2015;33:274–282. doi:10.3892/or.2014.3587
35. Zhou D, Luan J, Huang C, Li J. Tumor-associated macrophages in hepatocellular carcinoma: friend or foe? *Gut Liver*. 2020. doi:10.5009/gnl20223
36. Cannarile MA, Weisser M, Jacob W, Jegg AM, Ries CH, Ruttinger D. Colony-stimulating factor 1 receptor (CSF1R) inhibitors in cancer therapy. *J Immunother Cancer*. 2017;5:53. doi:10.1186/s40425-017-0257-y
37. Duan S, Guo W, Xu Z, et al. Natural killer group 2D receptor and its ligands in cancer immune escape. *Mol Cancer*. 2019;18:29. doi:10.1186/s12943-019-0956-8
38. Tai WT, Chu PY, Shiau CW, et al. STAT3 mediates regorafenib-induced apoptosis in hepatocellular carcinoma. *Clin Cancer Res*. 2014;20(22):5768–5776. doi:10.1158/1078-0432.CCR-14-0725
39. Uschner FE, Schueller F, Nikolova I, et al. The multikinase inhibitor regorafenib decreases angiogenesis and improves portal hypertension. *Oncotarget*. 2018;9(90):36220–36237. doi:10.18632/oncotarget.26333
40. Shigeta K, Matsui A, Kikuchi H, et al. Regorafenib combined with PD1 blockade increases CD8 T-cell infiltration by inducing CXCL10 expression in hepatocellular carcinoma. *J Immunother Cancer*. 2020;8:e001435. doi:10.1136/jitc-2020-001435
41. Raybould AL, Sanoff H. Combination antiangiogenic and immunotherapy for advanced hepatocellular carcinoma: evidence to date. *J Hepatocell Carcinoma*. 2020;7:133–142. doi:10.2147/JHC.S224938
42. da Fonseca LG, Reig M, Bruix J. Tyrosine kinase inhibitors and hepatocellular carcinoma. *Clin Liver Dis*. 2020;24(4):719–737. doi:10.1016/j.cld.2020.07.012
43. Strumberg D, Scheulen M, Schultheis B, et al. Regorafenib (BAY 73-4506) in advanced colorectal cancer: a Phase I study. *Br J Cancer*. 2012;106(11):1722–1727. doi:10.1038/bjc.2012.153
44. van Leeuwen RWF, van Gelder T, Mathijessen RHJ, Jansman FGA. Drug-drug interactions with tyrosine-kinase inhibitors: a clinical perspective. *Lancet Oncol*. 2014;15:e315–e326. doi:10.1016/S1470-2045(13)70579-5
45. Wilhelm SM, Carter C, Tang L, et al. BAY 43-9006 exhibits broad spectrum oral antitumor activity and targets the RAF/MEK/ERK pathway and receptor tyrosine kinases involved in tumor progression and angiogenesis. *Cancer Res*. 2004;64:7099–7109. doi:10.1158/0008-5472.CAN-04-1443
46. Bruix J, Tak WY, Gasbarrini A, et al. Regorafenib as second-line therapy for intermediate or advanced hepatocellular carcinoma: multicentre, open-label, phase II safety study. *Eur J Cancer*. 2013;49:3412–3419. doi:10.1016/j.ejca.2013.05.028
47. Bruix J, Merle P, Granito A, et al. Updated overall survival (OS) analysis from the international, Phase3, randomized, placebo-controlled RESORCE trial of regorafenib for patients with hepatocellular carcinoma (HCC) who progressed on sorafenib treatment. *Ann Oncol*. 2017;28:iii140. doi:10.1093/annonc/mdx262.008
48. Bruix J, Merle P, Granito A, et al. Comparison of modified (m) RECISTand RECIST 1.1 assessments in the phase 3 RESORCE trial comparing regorafenib and placebo in patients with hepatocellular carcinoma who progressed during sorafenib treatment. *J Hepatol*. 2017;66:S451–S452. doi:10.1016/S0168-8278(17)31281-3
49. Tovoli F, Renzulli M, Granito A, Golfieri R, Bolondi L. Radiologic criteria of response to systemic treatments for hepatocellular carcinoma. *Hepat Oncol*. 2017;4(4):129–137. doi:10.2217/hep-2017-0018
50. Bruix J, Merle P, Granito A, et al. Survival by pattern of tumor progression during prior sorafenib (SOR) treatment in patients with hepatocellular carcinoma (HCC) in the phase III RESORCE trial comparing second-line treatment with regorafenib (REG) or placebo. *J Clin Oncol*. 2017;35(4_suppl):229. doi:10.1200/JCO.2017.35.4_suppl.229
51. Bruix J, Merle P, Granito A, et al. Hand-foot skin reaction (HFSR) and overall survival (OS) in the phase 3 RESORCE trial of regorafenib for treatment of hepatocellular carcinoma (HCC) progressing on sorafenib. *J Clin Oncol*. 2018;36 (4_suppl):412. doi:10.1200/JCO.2018.36.4_suppl.412
52. Granito A, Marinelli S, Negrini G, Menetti S, Benevento F, Bolondi L. Prognostic significance of adverse events in patients with hepatocellular carcinoma treated with sorafenib. *Therap Adv Gastroenterol*. 2016;9:240–249. doi:10.1177/1756283X15618129
53. Finn RS, Merle P, Granito A, et al. Outcomes of sequential treatment with sorafenib followed by regorafenib for HCC: additional analyses from the phase III RESORCE trial. *J Hepatol*. 2018;69:353–358. doi:10.1016/j.jhep.2018.04.010

54. Observational study to evaluate, under real-world practice conditions, the safety and effectiveness of regorafenib in patients diagnosed with unresectable hepatocellular carcinoma (uHCC). Available from: https://clinicaltrials.gov/ct2/show/NCT03289273. Accessed April 20, 2021.
55. Merle P, Lim HY, Finn RS, et al. Sequential treatment with sorafenib followed by regorafenib in patients with unresectable hepatocellular carcinoma (HCC): interim analysis of the observational REFINE study. *J Clin Oncol*. 2020;38(15_suppl):e16680–e16680. doi:10.1200/JCO.2020.38.15_suppl.e16680
56. Yoo C, Park J-W, Kim YJ, et al. Multicenter retrospective analysis of the safety and efficacy of regorafenib after progression on sorafenib in Korean patients with hepatocellular carcinoma. *Invest New Drugs*. 2019;37:567–572. doi:10.1007/s10637-018-0707-5
57. Yoo C, Byeon S, Bang Y, et al. Regorafenib in previously treated advanced hepatocellular carcinoma: impact of prior immunotherapy and adverse events. *Liver Int*. 2020;40:2263–2271. doi:10.1111/liv.14496
58. Fung AS, Tam VC, Meyers DE, et al. Second-line treatment of hepatocellular carcinoma after sorafenib: characterizing treatments used over the past 10 years and real-world eligibility for cabozantinib, regorafenib, and ramucirumab. *Cancer Med*. 2020;9:4640–4647. doi:10.1002/cam4.3116
59. Kuzuya T, Uchikawa S, Kawaoka T, et al. Clinical outcomes of sorafenib treatment failure for advanced hepatocellular carcinoma and candidates for regorafenib treatment in real-world practice. *Hepatol Res*. 2018;48:814–820. doi:10.1111/hepr.13180
60. Granito A, Bolondi L. Non-transplant therapies for patients with hepatocellular carcinoma and Child-Pugh-Turcotte class B cirrhosis. *Lancet Oncol*. 2017;18:e101–e112. doi:10.1016/S1470-2045(16)30569-1
61. Ogasawara S, Chiba T, Ooka Y, et al. Characteristics of patients with sorafenib-treated advanced hepatocellular carcinoma eligible for second-line treatment. *Invest New Drugs*. 2018;36:332–339. doi:10.1007/s10637-017-0507-3
62. Terashima T, Yamashita T, Sunagozaka H, et al. Analysis of the liver functional reserve of patients with advanced hepatocellular carcinoma undergoing sorafenib treatment: prospects for regorafenib therapy. *Hepatol Res*. 2018;48:956–966. doi:10.1111/hepr.13196
63. Johnson PJ, Berhane S, Kagebayashi C, et al. Assessment of liver function in patients with hepatocellular carcinoma: a new evidence-based approach-the ALBI grade. *J Clin Oncol*. 2015;33:550–558. doi:10.1200/JCO.2014.57.9151
64. Yukimoto A, Hirooka M, Hiraoka A, et al. Using ALBI score at the start of sorafenib treatment to predict regorafenib treatment candidates in patients with hepatocellular carcinoma. *Jpn J Clin Oncol*. 2019;49:42–47. doi:10.1093/jjco/hyy153
65. Takada H, Kurosaki M, Tsuchiya K, et al. Baseline and early predictors of good patient candidates for second-line after sorafenib treatment in unresectable hepatocellular carcinoma. *Cancers*. 2019;11:1256. doi:10.3390/cancers11091256
66. Moriguchi M, Aramaki T, Sato R, et al. Intrahepatic tumor burden as a novel factor influencing the introduction of second-line chemotherapy for hepatocellular carcinoma. *Anticancer Res*. 2020;40:3953–3960. doi:10.21873/anticanres.14387
67. Kim HD, Bang Y, Lee MA, et al. Regorafenib in patients with advanced Child-Pugh B hepatocellular carcinoma: a multicentre retrospective study. *Liver Int*. 2020;40:2544–2552. doi:10.1111/liv.14573
68. Wang W, Tsuchiya K, Kurosaki M, et al. Sorafenib-regorafenib sequential therapy in Japanese patients with unresectable hepatocellular carcinoma-relative dose intensity and post-regorafenib therapies in real world practice. *Cancers (Basel)*. 2019;11:1517. doi:10.3390/cancers11101517
69. Iavarone M, Invernizzi F, Czauderna C, et al. Preliminary experience on safety of regorafenib after sorafenib failure in recurrent hepatocellular carcinoma after liver transplantation. *Am J Transplant*. 2019;19:3176–3184. doi:10.1111/ajt.15551
70. Teufel M, Seidel H, Köchert K, et al. Biomarkers associated with response to regorafenib in patients with hepatocellular carcinoma. *Gastroenterology*. 2019;156:1731–1741. doi:10.1053/j.gastro.2019.01.261
71. Jayson GC, Zhou C, Backen A, et al. Plasma Tie2 is a tumor vascular response biomarker for VEGF inhibitors in metastatic colorectal cancer. *Nat Commun*. 2018;9:4672. doi:10.1038/s41467-018-07174-1
72. Antoniotti C, Marmorino F, Pennati M, et al. Circulating angiogenesis-related markers as predictors of benefit from regorafenib in metastatic colorectal cancer (mCRC) patients (pts). *J Clin Oncol*. 2018;36(4_suppl):675. doi:10.1200/JCO.2018.36.4_suppl.675
73. Okusaka T, Ikeda M. Immunotherapy for hepatocellular carcinoma: current status and future perspectives. *ESMO Open*. 2018;3(Suppl 1):e000455. doi:10.1136/esmoopen-2018-000455
74. Liu JY, Chiang T, Liu CH, et al. Delivery of siRNA using CXCR4-targeted nanoparticles modulates tumor microenvironment and achieves a potent antitumor response in liver cancer. *Mol Ther*. 2015;23:1772–1782. doi:10.1038/mt.2015.147
75. Faivre S, Rimassa L, Finn RS. Molecular therapies for HCC: looking outside the box. *J Hepatol*. 2020;72:342–352. doi:10.1016/j.jhep.2019.09.010
76. Tsai AK, Khan AY, Worgo CE, Wang LL, Liang Y, Davila E. A multikinase and DNA-PK inhibitor combination immunomodulates melanomas, suppresses tumor progression, and enhances immunotherapies. *Cancer Immunol Res*. 2017;5:790–803. doi:10.1158/2326-6066.CIR-17-0009
77. Fukumura D, Kloepper J, Amoozgar Z, et al. Enhancing cancer immunotherapy using antiangiogenics: opportunities and challenges. *Nat Rev Clin Oncol*. 2018;15:325–340. doi:10.1038/nrclinonc.2018.29
78. Hamzah J, Jugold M, Kiessling F, et al. Vascular normalization in Rgs5-deficient tumours promotes immune destruction. *Nature*. 2008;453:410–414. doi:10.1038/nature06868
79. Jain RK. Antiangiogenesis strategies revisited: from starving tumors to alleviating hypoxia. *Cancer Cell*. 2014;26:605–622. doi:10.1016/j.ccell.2014.10.006
80. Huang Y, Yuan J, Righi E, et al. Vascular normalizing doses of antiangiogenic treatment reprogram the immunosuppressive tumor microenvironment and enhance immunotherapy. *Proc Natl Acad Sci U S A*. 2012;109:17561–17566. doi:10.1073/pnas.1215397109
81. Tian L, Goldstein A, Wang H, et al. Mutual regulation of tumor vessel normalization and immunostimulatory reprogramming. *Nature*. 2017;544:250–254. doi:10.1038/nature21724
82. Shigeta K, Datta M, Hato T, et al. Dual programmed death receptor-1 and vascular endothelial growth factor receptor-2 blockade promotes vascular normalization and enhances antitumor immune responses in hepatocellular carcinoma. *Hepatology*. 2020;71:1247–1261. doi:10.1002/hep.30889

83. Chen CW, Ou DL, Hsu CL, Lin L, Cheng AL, Hsu C. Regorafenib may enhance efficacy of anti-program cell death-1 (PD-1) therapy in hepatocellular carcinoma (HCC) through modulation of macrophage polarization. International Liver Congress. *J Hepatol*. 2019;70(suppl):e605–e606. doi:10.1016/S0618-8278(19)31207-1
84. Waldschmidt D, El-Khoueir AB, Kim RD, et al. Phase 1b study of regorafenib (REG) plus pembrolizumab (PEMBRO) for first-line treatment of advanced hepatocellular carcinoma (HCC). *Oncol Res Treat*. 2020;43(SUPPL 4):123.
85. Regorafenib Followed by Nivolumab in Patients With Hepatocellular Carcinoma (GOING). Available from: https://clinicaltrials.gov/ct2/show/NCT04170556. Accessed April 20, 2021.
86. Lee A, Keam SJ. Tislelizumab: first approval. *Drugs*. 2020;80:617–624. doi:10.1007/s40265-020-01286-z
87. Regorafenib plus tislelizumab as first-line systemic therapy for patients with advanced hepatocellular carcinoma. Available from: https://www.clinicaltrials.gov/ct2/show/NCT04183088?id=NCT04476329+OR+NCT04183088+OR+NCT04327700+OR+NCT04170556+OR+NCT02162914+OR+NCT04310709+OR+NCT03644511+OR+NCT03347292+OR+NCT01466504+OR+NCT03956940+OR+NCT03899428&draw=2&rank=4&load=cart. Accessed April 20, 2021.
88. A Phase I/II Study of regorafenib plus avelumab in solid tumors (REGOMUNE). Available from: https://clinicaltrials.gov/ct2/show/NCT03475953. Accessed April 20, 2021.
89. Tong M, Che N, Zhou L, et al. Efficacy of annexin A3 blockade in sensitizing hepatocellular carcinoma to sorafenib and regorafenib. *J Hepatol*. 2018;69:826–839. doi:10.1016/j.jhep.2018.05.034
90. Tutusaus A, Stefanovic M, Boix L, et al. Antiapoptotic BCL-2 proteins determine sorafenib/regorafenib resistance and BH3-mimetic efficacy in hepatocellular carcinoma. *Oncotarget*. 2018;9:16701–16717. doi:10.18632/oncotarget.24673
91. Li X, He G, Su F, et al. Regorafenib-loaded poly (lactide-co-glycolide) microspheres designed to improve transarterial chemoembolization therapy for hepatocellular carcinoma. *Asian J Pharm Sci*. 2020;15(6):739–751. doi:10.1016/j.ajps.2020.01.001
92. Feng M, Pan Y, Kong R, Shu S. Therapy of primary liver cancer. *Innovation (N Y)*. 2020;1:100032. doi:10.1016/j.xinn.2020.100032
93. McKay RR, Xie W, McGregor BA, et al. Optimized management of nivolumab (Nivo) and ipilimumab (Ipi) in advanced renal cell carcinoma (RCC): a response-based phase II study (OMNIVORE). *J Clin Oncol*. 2020;38(Suppl_15):5005. doi:10.1200/JCO.2020.38.15_suppl.5005
94. Atkins MB, Jegede O, Haas NB, et al. Phase II study of nivolumab and salvage nivolumab + ipilimumab in treatment-naïve patients with advanced renal cell carcinoma (RCC) (HCRN GU16-260). *J Clin Oncol*. 2020;38(Suppl_15):5006. doi:10.1200/JCO.2020.38.15_suppl.5006
95. Choueiri TK, Kluger HM, George S, et al. FRACTION-RCC: innovative, high-throughput assessment of nivolumab + ipilimumab for treatment-refractory advanced renal cell carcinoma (aRCC). *J Clin Oncol*. 2020;38(Suppl_15):5007. doi:10.1200/JCO.2020.38.15_suppl.5007
96. Choueiri TK, Albiges L, Powles T, et al. A phase 3 study (COSMIC-313) of cabozantinib in combination with nivolumab and ipilimumab in patients with previously untreated advanced renal cell carcinoma of intermediate or poor risk. Presented at: 2020 SITC Annual Meeting; November 9-14, 2020; Abstract 342.
97. Bekaii-Saab TS, Ou FS, Ahn DH, et al. Regorafenib dose-optimisation in patients with refractory metastatic colorectal cancer (ReDOS): a randomised, multicentre, open-label, phase 2 study. *Lancet Oncol*. 2019;20:1070–1082. doi:10.1016/S1470-2045(19)30272-4
98. Reiss KA, Yu S, Mamtani R, et al. Starting dose of sorafenib for the treatment of hepatocellular carcinoma: a retrospective, Multi-Institutional Study. *J Clin Oncol*. 2017;35:3575–3581. doi:10.1200/JCO.2017.73.8245
99. Iavarone M, Cabibbo G, Piscaglia F, et al. Field-practice study of sorafenib therapy for hepatocellular carcinoma: a prospective multicenter study in Italy. *Hepatology*. 2011;54:2055–2063. doi:10.1002/hep.24644
100. Facciorusso A, Abd El Aziz MA, Sacco R. Efficacy of regorafenib in hepatocellular carcinoma patients: a systematic review and meta-analysis. *Cancers (Basel)*. 2019;12:36. doi:10.3390/cancers12010036