European Journal of Cancer (2014) xxx, xxx–xxx

Available at www.sciencedirect.com

ScienceDirect

journal homepage: www.ejcancer.com

Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons

Antoine Hollebecque [a,b], David Malka [a], Charles Ferté [a], Michel Ducreux [a], Valérie Boige [a,*]

[a] Department of Cancer Medicine, Gustave Roussy, University of Paris Sud, Villejuif, France
[b] Drug Development Department (DITEP), Gustave Roussy, University of Paris Sud, Villejuif, France

Received 10 December 2014; accepted 11 December 2014

KEYWORDS
Hepatocellular carcinoma
Systemic treatment
Drug development
Molecular targeted agents
Signalling pathway
Molecular biomarkers
Sorafenib

Abstract Hepatocellular carcinoma (HCC) is an aggressive malignancy, which accounts for a third of all cancer deaths globally each year. The management of patients with HCC is complex, as both the tumour stage and any underlying liver disease must be considered conjointly. Since the approval of sorafenib in advanced HCC, several phase III clinical trials have failed to demonstrate any superiority over sorafenib in the frontline setting, and no agent has been shown to impact outcomes after sorafenib failure. This review will focus on the range of experimental therapeutics for patients with advanced HCC and highlight the successes and failures of these treatments as well as areas for future development. Specifics such as dose limiting toxicity and safety profile in patients with liver dysfunction related to the underlying chronic liver disease should be considered when developing therapies in HCC. Finally, robust validated and reproducible surrogate end-points as well as predictive biomarkers should be defined in future randomised trials.

0. Introduction

Hepatocellular carcinoma (HCC) is the third cause of cancer-related death worldwide [1]. The rim prognosis of HCC is in great part due to the fact that despite the implementation of screening programs targeting at-risk populations (i.e. patients with chronic liver disease) in most developed countries worldwide, many patients diagnosed with HCC (or HCC recurrence) are not amenable to curative-intent treatments. Despite numerous trials investigating various cytotoxic agents alone or in combination, the role of systemic chemotherapy in advanced HCC remains unclear. No drugs either alone or in combination have been shown to do better than doxorubicin, which did not convincingly improve survival over supportive care [2].

* *Corresponding author at:* Gustave Roussy, 114 rue Edouard Vaillant, F-94805 Villejuif, France. Tel.: +33 (0)142114311; fax: +33 (0)142115229.
E-mail address: Valerie.boige@gustaveroussy.fr (V. Boige).

http://dx.doi.org/10.1016/j.ejca.2014.12.005
0959-8049/

Indeed, in patients with advanced HCC, sorafenib, an orally available tyrosine kinase inhibitor (TKI) targeting – among others – vascular endothelial growth factor (VEGF), the key mediator of angiogenesis, and RAF, remains the only approved systemic therapy since the results of the two Phase III trials SHARP and Asia–Pacific [3,4] (Table 1). The efficacy of sorafenib in HCC is thought to result from the inhibition of VEGF and of the RAS/RAF/MEK/ERK pathway at the level of RAF. Irrespective of the mechanisms of action of sorafenib which remain not fully understood, the observed low objective response rate (ORR) according to response evaluation criteria in solid tumors (RECIST) (<5%) and the median overall survival (OS) of less than 1 year achieved in randomised studies emphasise the need for new treatments in HCC. This review highlights the results from phase three studies assessing molecular-targeting agents as first-line treatment in combination with, or compared to sorafenib, or as second-line therapy after failure of sorafenib, and details several drugs with new targets under evaluation in phase II and III trials as well as biomarker-driven therapeutic strategies.

1. Recent disillusions

1.1. Antiangiogenic agents

As high VEGF expression and increased micro-vessel density have been associated with poor survival, there is a strong rationale for using antiangiogenic agents in HCC [5,6]. BRISK-FL trial was based on the preclinical and promising clinical activity of brivanib, a dual TKI of

Table 1
Randomised phase III clinical trials completed in hepatocellular carcinoma (HCC) in the first- and second-line settings (2007–2014).

Comparison [Reference] (Name, study number)	Treatment line	Patients (*n*)	TTP (in months)	OS (in months)
Sorafenib versus placebo [3] (SHARP, NCT00105443)	1st	Sorafenib ($n = 299$) Placebo ($n = 303$)	5.5 versus 2.8; HR = 0.58 (95% CI, 0.45–0.74); $P < 0.001$	10.7 versus 7.9; HR = 0.69 (95% CI, 0.55–0.87); $P = 0.00058$
Sorafenib versus placebo [4] (Asia–Pacific, NCT00492752)	1st	Sorafenib ($n = 150$) Placebo ($n = 76$)	2.8 versus 1.4; HR = 0.57 (95% CI, 0.42–0.79); $P = 0.0005$	6.5 versus 4.2; HR = 0.68 (95% CI, 0.50–0.93); $P = 0.014$
Brivanib versus sorafenib [9] (BRISK-FL, NCT00858871)	1st	Brivanib ($n = 577$) Sorafenib ($n = 578$)	4.1 versus 4.2; HR = 1.01 (95% CI, 0.88–1.16); $P = 0.8$	9.5 versus 9.9; HR = 1.05 (95% CI, 0.94–1.23); $P = 0.31$
Sunitinib versus sorafenib [13] (SUN, NCT00247676)	1st	Sunitinib ($n = 530$) Sorafenib ($n = 544$)	3.8 versus 4.1; HR = 1.13 (95% CI, 0.98–1.31); $P = 0.16$	7.9 versus 10.2; HR = 1.30 (95% CI, 1.13–1.5); $P = 0.001$
Linifanib versus sorafenib [14] (LIGHT, NCT01009593)	1st	Linifanib ($n = 517$) Sorafenib ($n = 518$)	5.4 versus 4.0; HR = 0.76 (95% CI, 0.64–0.89); $P < 0.001$	9.1 versus 9.8; HR = 1.04 (95% CI, 0.89–1.22); $P =$ NS
Ramucirumab versus placebo [17] (REACH, NCT01140347)	2nd	Ramucirumab ($n = 283$) Placebo ($n = 282$)	3.5 versus 2.6; HR = 0.59 (95% CI, 0.49–0.72); $P = 0.0001$	9.2 versus 7.6; HR = 0.866 (95% CI, 0.72–1.05); $P = 0.14$
Brivanib versus placebo [18] (BRISK-PS, NCT01108705)	2nd	Brivanib ($n = 263$) Placebo ($n = 132$)	4.2 versus 2.7; HR = 0.56 (95% CI, 0.42–0.78); $P = 0.001$	9.4 versus 8.2; HR = 0.89 (95% CI, 0.69–1.15); $P = 0.33$
FOLFOX versus doxorubicin [24] (NCT00471965)	1st	FOLFOX ($n = 184$) Doxorubicin ($n = 187$)	2.93 versus 1.77 (95% CI, 1.6–2.3)*; $P = 001$	6.4 versus 4.9; HR = 0.80 (95% CI, 0.63–1.02); $P = 0.07$
Everolimus versus placebo [47] (EVOLVE-1, NCT01035229)	2nd	Everolimus ($n = 362$) Placebo ($n = 184$)	3.0 versus 2.6; HR = 0.93 (95% CI, 0.75–1.15); P: NA	7.6 versus 7.3; HR = 1.05 (95% CI, 0.86–1.27); $P = 0.67$
Sorafenib + erlotinib versus sorafenib + placebo [52] (SEARCH, NCT00901901)	1st	Sorafenib + erlotinib ($n = 362$) Sorafenib + placebo ($n = 358$)	3.2 versus 4.0; HR = 1.13 (95% CI, 0.94–1.36); $P = 0.91$	9.5 versus 8.5; HR = 0.92 (95% CI, 0.78–1.1); $P = 0.2$

Mo = months; HR = hazard ratio; ns = not significant; OS = overall survival; PFS = progression-free survival; TTP = time to progression; CI = confidence interval.
* PFS.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

VEGF receptors (VEGFR) and fibroblast growth factor receptors (FGFR) [7,8]. This phase III trial failed to demonstrate either superiority or non-inferiority over sorafenib in 1150 patients [9]. In the two groups, similar results were observed for median OS, ORR, time to progression (TTP) and disease control rates (DCR) (Table 1). Toxicity and proportion of patients discontinuing therapy for toxicity seemed somewhat higher in the brivanib arm. The BRISK-PS trial compared brivanib to placebo (with 2:1 randomisation) after failure of sorafenib in 395 patients in Western centres. Despite significant improvements in ORR (10% versus 2%; $P = 0.003$) as measured by modified RECIST and in TTP, BRISK-PS failed to achieve its primary end-point of a significant improvement in OS (Table 1). Imbalances between the treatment arms regarding macro-vessel invasion and baseline alphafetoprotein (AFP) levels favouring the placebo arm, the higher rates of discontinuation for toxicity in the brivanib arm, and the higher-than-expected OS of 8.2 months in the placebo arm reflecting the highly selected nature of the population may explain these negative results. A randomised phase II trial comparing axitinib, a potent and selective TKI of VEGFR-1, -2 and -3 to placebo in 202 patients after failure of sorafenib failed to demonstrate a significant benefit in OS, its primary end-point) (12.7 versus 9.7 months; hazard ratio (HR) = 0.87; $P = 0.211$) despite significantly better progression-free survival (PFS) and DCR [10]. Sunitinib, a multi-targeted TKI has shown anti-tumour activity among HCC patients in several single-arm phase II studies [11,12]. SUN, a large phase III trial comparing sunitinib (37.5 mg daily) to sorafenib in the first-line setting, was discontinued early after the inclusion of 1074 patients because of higher toxicity (grade 3–4 adverse events, 82% versus 74%), including more grade 3–5 bleeding events, and shorter OS [13] (Table 1). Linifanib is the third selective TKI of VEGFR and platelet-derived growth factor receptor (PDGFR) compared to sorafenib in a phase III trial in the first-line setting [14]. After the inclusion of 1035 patients, median OS was similar despite median TTP and ORR (13.0 versus 6.9%) in favour of linifanib (Table 1). Again, toxicity leading to treatment discontinuations and dose reductions was significantly more frequent with linifanib.

The distinct safety profile of monoclonal antibodies targeting VEGF or VEGFR compared to TKI might be of special interest in patients with HCC. However, despite encouraging signals of activity with bevacizumab, a monoclonal antibody that binds VEGF-A, the main isoform of circulating VEGF, and notably interesting ORR of 13% and 14%, as first-line single-agent therapy in two Phase II studies [15,16], the development of this agent was stopped in HCC because of safety concerns (infrequent gastro-oesophageal variceal bleeding). Moreover, in the REACH phase III trial, which allocated 565 patients to receive either ramucirumab, a fully human IgG1 monoclonal antibody targeting VEGFR-2, or placebo every 2 weeks after failure of sorafenib [17], arterial hypertension, liver dysfunction, bleeding and cytopaenias were more frequent in the ramucirumab arm, and despite significant benefit in terms of PFS, TTP, ORR and DCR, no significant OS benefit was seen, except in patients with AFP levels >400 ng/mL (7.8 versus 4.2 months; HR = 0.67; $P = 0.0059$) (Table 1). In this subgroup, which accounts for nearly half of the patients, an interesting ORR of 20% was achieved, suggesting a potential for further investigation of ramucirumab in such patients. However, whether baseline AFP levels are able to predict the efficacy of ramucirumab should be assessed prospectively, and the biological reasons underlying such a relationship (if any) remain elusive. More generally, owing to the repeatedly disappointing results observed in all recent phase III trials with antiangiogenic TKI as well as monoclonal antibodies, in both the first-line and the second-line setting, it seems unlikely that any of the numerous antiangiogenic agents still under evaluation among phase I/II (axitinib, AMG386, apatinib, dalantercept, nintedanib, orantinib, RO5323441, tivozanib and TRC-105) and randomised phase II-III trials (dovitinib, lenvatinib and regorafenib) [18–20] (Table 2) will be successful in the treatment of unselected patients with advanced HCC, even those with promising efficacy results in Phase II trials (e.g. lenvatinib, with median TTP and OS of 12.8 and 18.7 months, respectively [21]). Unfortunately, no biomarker predictive of the efficacy of antiangiogenic therapy in HCC has been validated so far, precluding the selection of patients in future trials [22,23].

1.2. Cytotoxic chemotherapy

The oxaliplatin-fluorouracil combination (FOLFOX regimen) has been recently compared to doxorubicin in a phase III trial. In the EACH Asian trial in which 371 patients were randomly assigned to FOLFOX or doxorubicin as first-line systemic treatment [24], ORR (8.2% versus 2.7%) and median PFS were significantly higher in the FOLFOX arm; however, the observed OS benefit attained statistical significance only in a subsequent post-hoc analysis (6.5 versus 4.9 months; $P = 0.04$) (Table 1). Whether oxaliplatin-based chemotherapy could represent a therapeutic option after failure of sorafenib deserves to be assessed prospectively.

1.3. Combination therapies with sorafenib

1.3.1. Combination of sorafenib with cytotoxic chemotherapy

Given the low ORR obtained with VEGFR inhibitors such as sorafenib (<3%), adding cytotoxic chemotherapy

Table 2
Main ongoing randomised trials.

Comparison (Name, study number)	Trial phase	Line of treatment	Primary end-point	Child–Pugh max	ECOG Max
Lenvatinib versus sorafenib (NCT01761266)	III	1st	OS	A	1
Sorafenib + doxorubicin versus sorafenib (CALGB80802, NCT01015833)	III	1st	OS	A	1
Regorafenib versus placebo (NCT01774344)	III	2nd	OS	A	1
Tivantinib versus placebo (Metiv-HCC, NCT01755767)	III	2nd	OS	A	1
Cabozantinib versus placebo (CELESTIAL, NCT01908426)	III	2nd	OS	A	1
ADI-PEG20 versus placebo (NCT01287585)	III	2nd	OS	B7	2
Dovitinib versus sorafenib (NCT01232296)	IIR	1st	OS	A	1
JX-594 versus BSC (NCT01387555)	IIR	2nd	OS	B7	2

OS = overall survival; PFS = progression-free survival; TTP = time to progression; MTD = maximum tolerated dose; NA = non-available; R = randomised trial; BSC = best supportive care.

to sorafenib might bring clinical value. In a randomised phase II study of doxorubicin with or without sorafenib in 96 patients, median OS, PFS (6.0 versus 2.7 months) and TTP were significantly improved in the sorafenib arm [25] (Table 3). However, whether this benefit is due to sorafenib by itself or to its combination to doxorubicin cannot be claimed before the final results of the on-going phase III trial comparing sorafenib with or without doxorubicin (CALGB 80802 trial) (Table 2).

Previous several first-line phase II studies of gemcitabine plus oxaliplatin combination (GEMOX) in advanced HCC showed a favourable safety profile, and encouraging ORR and DCR of 20% and 65%, respectively [26,27]. These results have recently been confirmed in 'real life' conditions and also as second-line treatment after failure of sorafenib in two retrospective studies [28,29]. Therefore, a randomised phase II trial was conducted in 94 patients allocated to either sorafenib alone or sorafenib plus GEMOX [30]. The main objective (4-month PFS rate >50%) was achieved but was comparable in both arms (54% versus 64% respectively), and median PFS and OS were not significantly different between the two arms (Table 3). In the SECOX single-arm phase II trial, sorafenib was combined to oxaliplatin (85 mg/m^2, day 1) and capecitabine (1700 mg/m^2, day 1–7) every 2 weeks. Among the 51 patients enrolled (84% of whom being hepatitis B carriers), 16% achieved an OR and another 62% had a stable disease for at least eight weeks [31]. However, median TTP and OS did not seem very different from those obtained with sorafenib alone (Table 3). Collectively, these data suggest that the addition of cytotoxic chemotherapy to sorafenib results at most in a modest synergism/additive effect.

1.3.2. Combination of sorafenib with EGFR inhibitors

Epidermal Growth Factor Receptor (EGFR) protein overexpression and EGFR gene amplification have been reported in 66% and 45% of HCC cases, respectively [32]. Pre-clinical data have also suggested a synergism between sorafenib and EGFR inhibitors as well as a role for the EGFR pathway in resistance to sorafenib [33,34]. However, the phase II studies of EGFR inhibitors administered either alone or in combination with cytotoxic chemotherapy or other molecular targeting agents reported to date failed to demonstrate significant or reproducible antitumour activity in advanced HCC [35–44]. Despite the lack of phase II data regarding the combination of sorafenib plus erlotinib in HCC, the SEARCH phase III trial was conducted, and concluded that the addition of erlotinib to sorafenib did not confer any survival benefit [45] (Table 1). Among potential explanations for these negative results, one includes the fact that adding erlotinib led to cumulative AEs decreasing sorafenib treatment duration.

1.3.3. Combination of sorafenib with mTOR inhibitors

Approximately 40–50% of the patients with HCC harbour tumour alterations in the mTOR signalling pathway [46]. Everolimus, an orally available mTOR

Table 3
Main clinical trials combining sorafenib and systemic chemotherapy.

Comparison [Reference] (Name, study number)	Trial Phase	Line of treatment	Patients (*n*)	TTP (in months)	OS (in months)
Doxorubicin + sorafenib versus doxorubicin + placebo [25] (NCT00108953)	II	1st	47 versus 49	6.4 versus 2.8; HR = 0.50 (95% CI, 0.3–0.9); $P = 0.02$	13.7 versus 6.5; HR = 0.49 (95% CI, 0.3–0.8); $P = 0.006$
GEMOX + sorafenib versus sorafenib [30] (GONEXT, NCT00941967)	II	1st	39 versus 44	6.2 versus 4.6* HR = NA; $P = 0.68$	13.5 versus 13.0 HR = NA; $P = 0.11$
Sorafenib + CAPOX [31] (SECOX, NCT00752063)	II	1st	51	5.3 (95% CI, 3.8–5.9)	11.7 (95% CI, 8.9–15.4)

GEMOX = gemcitabine plus oxaliplatin; CAPOX = capecitabine plus oxaliplatin; HR = hazard ratio; CI = confidence interval.
* PFS.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

inhibitor, failed to demonstrate a significant benefit over placebo in the phase III EVOLVE-1 trial in 546 patients who previously failed sorafenib [47] (Table 1). Temsirolimus, an intravenously administered mTOR inhibitor, met its primary end-point with a median PFS of 17 weeks and a 24-week PFS rate of 37% in a phase II study in 25 patients [48]. However, it failed to reproduce such encouraging results in three separate phase I–II trials, when it was used in monotherapy or in combination with sorafenib or bevacizumab [49–51]. As no predictive marker of efficacy is currently able to predict mTOR inhibitor efficacy, the development of mTOR inhibitors in HCC seems jeopardised [52].

2. New therapeutic agents on the horizon

2.1. MET inhibitors

Accumulating evidence has established the role of the tyrosine kinase receptor MET and its ligand hepatocyte growth factor (HGF) in tumour development and metastatic progression in HCC [53–55]. The activation of the HGF/MET pathway in HCC is associated with an aggressive phenotype and poor prognosis [56]. In a randomised phase II study, tivantinib, a selective MET TKI, has been compared to placebo in the second-line setting in 107 patients, of whom 77 (72%) could be assessed for tumour MET status by immunochemistry [57]. Of those, 37 (48%) had MET-high tumours. Overall, the 71 patients assigned to tivantinib had a slightly longer TTP (1.6 versus 1.4 months (HR 0.64, $P = 0.04$). The most frequent adverse event leading to dose reductions in the tivantinib group was severe neutropenia in eight (47%) of 17 patients, suggesting an off-target effect of this drug. Recent preclinical data also showed that tivantinib inhibits microtubule polymerisation in addition to inhibiting c-MET [58].

Interestingly, only patients with high tumour MET expression derived a significant benefit from tivantinib in terms of TTP (2.7 versus 1.4 months; HR = 0.43; $P = 0.03$) and OS (7.2 versus 3.8 months; HR = 0.38; $P = 0.01$) [57]. In the placebo group, patients with MET-high tumours had significantly shorter OS (3.8 months) compared to that in patients with MET-low tumours (9.0 months; HR = 2.94; $P = 0.02$). Therefore, as in other tumour types, tumour MET status appears both predictive and prognostic. For the first time in advanced HCC, the use of such a potential predictive biomarker has been showed to select patients who are most likely to benefit from treatment. However, MET status determination needs further standardisation and validation before being used routinely. A phase III trial of tivantinib in the second-line setting in patients with MET-high advanced HCC who previously failed sorafenib is currently in progress (Table 2). Cabozantinib (XL-184), a dual MET/VEGFR-2 TKI, has also shown encouraging results as a second-line therapy in a randomised phase II study in 41 patients, with a 12-week DCR of 68%, a median PFS of 4.4 months and a median OS of 15.1 months [59]. Three patients (8%) experienced a partial response, and 28 of the 36 evaluable patients (78%) had tumour regression. Most common grade 3/4 events were diarrhoea (17%), palmar-plantar erythrodysesthesia (15%) and thrombocytopenia (10%). Cabozantinib has entered in phase III (Table 2). Many other MET inhibitors (e.g. foretinib, golvantinib, INC-280 and MSC2156119J) are also under evaluation [60,61]. A plasma biomarker analysis from the SHARP trial supports the theory that a low HGF concentration at the baseline was a predictor of outcome in patients with HCC (P of interaction = 0.073) [62]. If this association is confirmed, this predictor could increase the scope of MET inhibitors in HCC.

2.2. Immune-based therapies

2.2.1. Immune checkpoint blockers

Immunotherapy aims to provide an efficient and selective targeting of tumour cells by inducing or boosting the existing tumour-specific immune response [63]. Blockade of immune checkpoints is one of the most promising approaches to activate anti-tumour immunity [64]. In advanced melanoma, ipilimumab, a monoclonal antibody directed against cytotoxic T-lymphocyte-associated antigen 4 (CTLA4), has demonstrated a survival benefit [65,66]. These results pioneered the use of immune checkpoint blockers in cancer. Owing to a microenvironment rich in immune cells and a high gene mutation rate which might predict tumour response to immunotherapy, HCC might be a good candidate for immune-based therapies [67–69]. In a pilot study in 21 patients with HCC and chronic hepatitis C infection, tremelimumab, an anti-CTLA4 monoclonal antibody, showed very promising ORR and DCR of 18% and 76%, respectively. Moreover, tremelimumab induced a significant decrease in viral load [70]. Despite safety concerns of this therapeutic class, particularly immune events, tremelimumab showed a good safety profile in this small study.

Programed death-1 (PD-1) is another immune checkpoint receptor that inhibits T-cell activation when bound by PD ligands (PDL)-1 and PDL-2. Very encouraging results in various tumour types have recently been published [71–73]. With reported overexpression rates varying from 45% to 93%, PDL-1 represents a promising target in HCC [74,75]. Preclinical data have suggested that PD-1 and PDL-1 can suppress HCC growth [76]. Nivolumab, a PD-1 blocking antibody is currently tested in a phase I trial dedicated to HCC patients. At the same time, several phase I studies of PD-1 and PDL-1 blockers opened specific cohorts for HCC patients [77,78]. In addition, these agents will be investigated as antiviral drugs and for earlier stages of

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

disease within the context of percutaneous tumour ablation and transarterial chemoembolisation.

2.2.2. Viral therapies

JX-594, an oncolytic and immunotherapeutic vaccina virus, has recently been evaluated in a phase II trial in which patients were randomised to receive low- or high-dose of the product by direct intratumour injection [79]. The intrahepatic 8-week DCR was 46% and high dose level of JX-594 was associated with a longer OS compared to low dose. Randomised controlled trials are ongoing comparing JX-594 versus standard of care in the first-line setting and versus best supportive care after sorafenib failure.

2.2.3. Lenalidomide

Lenalidomide is a thalidomide analogue with immunomodulatory and antiangiogenic properties that include altering cytokine production, activating T cells and stimulating natural killer cell functions [80]. Lenalidomide has been tested in a phase II trial in patients with advanced HCC who failed sorafenib. Six (15%) of the 40 patients had a partial tumour response. Two patients (5%) did not progress at 36 and 32 months. The median PFS and OS were 3.6 and 7.6 months, respectively.

2.3. MEK inhibitors

In a phase II study, selumetinib (AZD6244), an orally available MEK inhibitor produced no response in the 17 evaluable patients, and the study was stopped at the interim analysis. Median TTP was short (8 weeks) suggesting minimal clinical activity [81]. Selumetinib has also been tested in combination with sorafenib. Although three partial responses and six disease stabilisations were observed among the 11 evaluable patients, the small number of patients renders this study difficult to interpret [82].

Refametinib (BAY 86-9766), another orally available MEK inhibitor, has been tested in combination with sorafenib in the first-line setting [83]. Encouraging efficacy results have been observed among the 65 evaluable patients with a 43% DCR (ORR, 5%). Best clinical responders had *RAS* mutations. However, many concerns were raised regarding toxicity, including four fatal adverse events (hepatic failure, sepsis/hepatic encephalopathy, tumour lysis syndrome and unknown cause). The most frequent drug-related adverse events were skin rash, gastrointestinal toxicity (nausea, vomiting and anorexia) and elevation of aminotransferases. Dose modifications due to adverse events were necessary in almost all patients. The frequency and severity of the drug-related adverse events among the different clinical trials may hamper the development of MEK inhibitors in HCC. A phase II single-agent study is ongoing in patients with advanced HCC carrying a *RAS* mutation.

2.4. Telomerase inhibitors

Telomeres are protective caps at the ends of human chromosomes. Telomeres shorten with each successive cell division in normal human cells whereas they are continuously elongated by human telomerase reverse transcriptase (hTERT) in tumours [84]. TERT promoter mutations are highly related to the stepwise hepatocarcinogenesis, and mutations are identified in 42% of HCC (Table 4) [85]. Several strategies of direct or indirect telomerase inhibition (antisense oligonucleotides, immunotherapy, gene therapy, G-quadruplex stabilisers, telomere and telomerase-associated proteins (HSP90)...) are under investigation. Most of these agents have entered phase I and II clinical trials in patients with various tumours. Some of them have shown signs of antitumour activity but most therapeutics have shown to be more effective when used in combination with standard therapies. Further data are needed to confirm the role of this therapeutic class.

2.5. FGF inhibitors

FGF19, the natural ligand of FGFR4, is activated in approximately 20% of HCC [86–88]. As promising clinical data using selective FGFR inhibitors have already been reported among various tumour types with FGFR amplification, FGFR4 blockers could represent an interesting approach [89].

Table 4
Main molecular alterations in HCC.

Pathway [117,118]	Mutated gene	Estimated frequency (%)
Telomere maintenance	*TERT*	32
Wnt/β-catenin	*CTNNB1*	19–32.8
	AXIN1	11
	APC	1.6
Cell cycle regulator	*TP53*	20.8–30
	CDKN2A	7.2–9
	ATM	3
	IRF2	4.8
	RB1	3
Epigenetic modifier	*MLL genes*	3
	ARID1A	9–16.8
	ARID2	5.6–7
	SMARCA4	3
FGF/PIK3/Ras signalling	*FGF19*[*]	20
	KRAS	1.6
	NRAS	2
	BRAF	3
	PTEN	3
	PIK3CA	5
	RPS6KA3	9.6
	EGFR	<1
Stress oxidative pathway	*NFE2L2*	3–6.4
	BRCA2	2
JAK/STAT pathway	*JAK1*	0–9
	IL6ST	5
Other	*HNF1A*	5

[*] Amplification.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

2.6. IGF1-R inhibitors

To date, insulin-like growth factor (IGF)1-R inhibitor development seems compromised, as several drugs (AVE-1642, BIIB 022, cixutumumab, OSI-906) showed a lack of antitumour activity in phase I/II trials in HCC [90,91].

2.7. Apoptosis inducers

Tumour necrosis factor-related apoptosis-inducing ligand (TRAIL) is a member of the tumour necrosis factor ligand super family that induces programed cell death primarily in tumour cells through TRAIL death receptors. Mapatumumab, a TRAIL-R1 agonistic antibody, has been tested in combination with sorafenib in the first-line setting. In this phase II study in which 101 patients were randomised to receive sorafenib with either mapatumumab or placebo, no PFS or OS improvement was observed [92].

2.8. Glypican-3

Glypican-3, a member of heparan sulphate proteoglycan family, is highly expressed in HCC [93,94]. GC33, a humanised monoclonal antibody that binds the glypican-3 receptor, interacts with CD16/FcγR3 and triggers antibody-dependent cytotoxicity. In a phase II trial in patients with advanced HCC who had failed prior systemic therapy, GC33 did not show any clinical benefit compared to placebo [95,96]. Attempts to combine GC33 to sorafenib led to very poor tolerance [97].

2.9. TGF-β inhibitors

Transforming Growth Factor-beta (TGF-β) is the most important profibrogenic mediator and a protumourigenic factor [98]. In a phase II trial, LY2157299, a TGF-β inhibitor, was administered to 109 patients at two different dose levels [99]. In the whole patient population, median TTP and OS were 12 and 36 weeks, respectively. In AFP responders (AFP decline >20%), median OS was 93.1 weeks versus 29.6 weeks in AFP non-responders. The safety profile appeared manageable, with only four patients who discontinued treatment due to a drug-related adverse event. Combination with sorafenib is under evaluation.

2.10. Arginine deprivation

HCC is an arginine auxotroph due to argininosuccinate synthetase I deficiency and extracts arginine from the circulation [100]. Arginine deprivation induces cell death [100,101]. Several phase I/II trials have tested different arginine deprivation therapies (ADI-PEG 20, Peg-rhArg1) [102–105]. A phase I/II study of pegylated arginine deiminase (ADI-PEG 20), an arginine-degrading enzyme, showed a favourable safety profile in a patient population with impaired hepatic function [106]. Of 19 evaluable patients, two (10%) had a complete response, 7 (37%) had a partial response and 7 (37%) had stable disease. In an Asian randomised phase II trial, ADI-PEG 20 showed a promising DCR of 31%, a median OS of 7.3 months and an excellent toxicity profile [102]. Based on these results, a double-blind, placebo-controlled, phase III study of ADI-PEG 20 versus best supportive care after prior systemic therapy is ongoing (Table 2).

2.11. Histone deacetylase inhibitors

Histone deacetylase (HDAC) inhibitors modulate the expression of genes by causing an increase in histone acetylation. Among the HDAC inhibitors already tested, belinostat showed disappointing results as single-agent therapy [107]. However, the combination of sorafenib with resminostat achieved an unexpectedly high 12-week PFS rate of 70% and a median OS of 8.0 months in a randomised phase II study (SHELTER) [108]. The future of this therapeutic class in HCC probably lies in combination therapy.

3. What can we change?

3.1. Clinical trial design

Compared to renal cell carcinoma or melanoma, the drug approval rate of new drugs in HCC has been particularly low. Recently, seven phase III trials failed to meet their primary end-points in the first-line (sunitinib, linifanib, brivanib and erlotinib) and second-line setting (brivanib, everolimus and ramucirumab). The main reasons explaining these failures have been extensively depicted recently [109]: (1) suboptimal understanding of HCC tumour drivers and molecular subclasses; (2) inappropriate determination of the maximum tolerated dose among HCC patients in the context of cirrhosis and liver dysfunction; (3) difficult-to-interpret non-randomised phase II trial design using inconsistent surrogate end-points and (4) lack of useful biomarkers. Thus, specific rigorous phase 1 clinical trial development is required. Liver tumour biopsy needs to be performed in all HCC patients eligible for systemic therapy to consider the molecular intertumoural heterogeneity of this disease and to facilitate predictive biomarker identification. Phase II development should define reliable surrogates of OS, including improved radiologic criteria to measure tumour shrinkage/stabilisation, as current criteria (RECIST, modified RECIST, ratio of tumour volume to necrosis…) are still not validated yet.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

3.2. From molecular data to predictive biomarkers

Different international collaborative studies such as The Cancer Genome Atlas (TCGA) and the International Cancer Genome Consortium (ICGC) have performed complete genomic characterisation of various cancers including HCC to refine our knowledge of the mutational landscape and the related signalling pathways involved in liver carcinogenesis [69]. Cancer genomes sequenced so far have revealed hundreds to thousands of mutations [110–114]. Biology of HCC is also complex, and no single oncogenic addiction loop has been found that drives tumour hepatocarcinogenesis and progression in HCC [69,115–118]. The most frequent HCC-related molecular alterations involved in HCC are summarised in Table 4. The use of predictive biomarkers has become essential leading to successful translational and clinical development of agents such as vemurafenib, crizotinib and olaparib [119–121]. Recent negative results from phase III trials highlighted the need to use such predictive biomarkers to improve the drug approval rate. Indeed, the identification of new relevant targets will lead to the development of pivotal proof-of-principle, proof-of-concept and biomarker-based enrichment trials [122–124]. However, such predictive biomarkers are not easy to find, and none has been validated so far in HCC. In a retrospective study looking at ten potential predictors of response to sorafenib, none of them were found to be predictive [62]. Recently, promising data emerged from MET and VEGF pathways. MET immunochemical expression has been suggested as a useful predictive biomarker to select patients who are most likely to benefit from the MET inhibitor tivantinib. VEGF-A amplification has retrospectively been described as a potential predictor of benefit for VEGF-A blocking drugs. FISH-based selection of VEGF-A-amplified HCC defined a group of sorafenib-treated patients with improved outcome [22,23,125]. Further prospective studies are required to validate these predictive biomarkers.

To speed up the drug development process, different initiatives worldwide have incorporated a molecular screening approach in order to match patients with a specific genomic alteration to the most relevant targeted therapy [126], and have demonstrated the feasibility of such an approach. However, several limitations might be stressed. The first one is linked to the interpretation of the sequencing results and to the selection of actionable targets for targeted therapies. Among the hundreds to thousands of mutations, making the distinction between driver mutations, which confer growth advantage and are responsible for pathogenesis, and 'bystander' mutations which are generally distributed randomly across the genome, is crucial to determine which one has to be targeted [127]. The second challenge is to take into account the intra-patient tumour heterogeneity and clonal evolution which can challenge patient selection and may explain the emergence of drug resistance. In HCC, a study suggested that among patients with multiple tumour nodules, 36% of them had tumours with different clonality and hence were of multicentric origin [128]. The third challenge is that the efficacy of targeted therapies is limited by drug resistance [129–131]. Sequential molecular evaluation would then be necessary to identify the mechanism of resistance [132,133]. New surrogate molecular markers derived from circulating tumour cells or cell-free DNA would then be of great interest to avoid tumour biopsies [134–137].

4. Conclusion

Since the approval of sorafenib, HCC remains deprived of additional systemic treatments in first and second-line therapy. The failure of all recent phase III trials has highlighted the complexity of drug development in HCC. Regardless of treatment, liver dysfunction, ethnic origin and the cause of liver disease should be increasingly taken into account. Robust, validated and reproducible surrogate end-points as well as predictive biomarkers of drug efficacy need to be defined and implemented in trial design. Finally, molecular complexity and heterogeneity of HCC including multiple genetic and epigenetic alterations should lead to identify distinct patient subgroups and hopefully improve patient outcome. In this perspective, novel trials should be designed to test drugs in biomarker-based HCC patient subpopulations.

Conflict of interest statement

DM has consulted or advised for Roche, Amgen, Imclone, Bayer, Teva, Keocyt, Sanofi-Aventis and Boehringer Ingelheim; has received honoraria from Merck Serono, Ipsen, Celgene and Novartis; has received travel grants from Roche and Bayer and has received research funding from Institut National du Cancer (INCa), Merck Serono, Amgen, Sanofi-Aventis and Roche.

MDu has consulted or advised for Roche, Merck Serono, Amgen, Lilly, Keocyt, Sanofi-Aventis, Ipsen, Celgene, Boehringer Ingelheim and Novartis; has received honoraria from Roche, Amgen, Merck Serono, Lilly and Novartis; has received travel grants from Roche, Amgen, Merck Serono and Novartis and has received research funding from Roche.

VB has consulted or advised for Merck Serono, Amgen, Sanofi-Aventis and Bayer; has received honoraria from Amgen, Merck Serono and Bayer; has received travel grants from Amgen, Merck Serono and Sanofi and has received research funding from Merck Serono.

CF and AH: None.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

References

[1] Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin 2011;61(2):69–90.
[2] Lai CL, Wu PC, Chan GC, Lok AS, Lin HJ. Doxorubicin versus no antitumor therapy in inoperable hepatocellular carcinoma. A prospective randomized trial. Cancer 1988;62(3):479–83.
[3] Llovet JM, Ricci S, Mazzaferro V, Hilgard P, Gane E, Blanc JF, et al. Sorafenib in advanced hepatocellular carcinoma. N Engl J Med 2008;359(4):378–90.
[4] Cheng AL, Kang YK, Chen Z, Tsao CJ, Qin S, Kim JS, 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(1):25–34.
[5] Chao Y, Li CP, Chau GY, Chen CP, King KL, Lui WY, et al. Prognostic significance of vascular endothelial growth factor, basic fibroblast growth factor, and angiogenin in patients with resectable hepatocellular carcinoma after surgery. Ann Surg Oncol 2003;10(4):355–62.
[6] Jeng KS, Sheen IS, Wang YC, Gu SL, Chu CM, Shih SC, et al. Prognostic significance of preoperative circulating vascular endothelial growth factor messenger RNA expression in resectable hepatocellular carcinoma: a prospective study. World J Gastroenterol 2004;10(5):643–8.
[7] Huynh H, Ngo VC, Fargnoli J, Ayers M, Soo KC, Koong HN, 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(19):6146–53.
[8] Park JW, Finn RS, Kim JS, Karwal M, Li RK, Ismail F, et al. Phase II, open-label study of brivanib as first-line therapy in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2011;17(7):1973–83.
[9] Johnson PJ, Qin S, Park JW, Poon RT, Raoul JL, Philip PA, et al. Brivanib versus sorafenib as first-line therapy in patients with unresectable, advanced hepatocellular carcinoma: results from the randomized phase III BRISK-FL study. J Clin Oncol 2013;31(28):3.
[10] Kang Y, Yau T, Park J, Boucher E, Lim H, Poon R, et al. Randomised study of axitinib plus best supportive care versus placebo plus BSC in patients with advanced hepatocellular carcinoma following prior antiangiogenic therapy. ESMO Congress 2014 2014;17.
[11] Faivre S, Raymond E, Boucher E, Douillard J, Lim HY, Kim JS, et al. Safety and efficacy of sunitinib in patients with advanced hepatocellular carcinoma: an open-label, multicentre, phase II study. Lancet Oncol 2009;10(8):794–800.
[12] Koeberle D, Montemurro M, Samaras P, Majno P, Simcock M, Limacher A, et al. Continuous Sunitinib treatment in patients with advanced hepatocellular carcinoma: a Swiss Group for Clinical Cancer Research (SAKK) and Swiss Association for the Study of the Liver (SASL) multicenter phase II trial (SAKK 77/06). Oncologist 2010;15(3):285–92.
[13] Cheng AL, Kang YK, Lin DY, Park JW, Kudo M, Qin S, et al. Sunitinib versus sorafenib in advanced hepatocellular cancer: results of a randomized phase III trial. J Clin Oncol 2013;31(32):4067–75.
[14] Cainap C, Qin S, Huang W-T, Chung I-J, Pan H, Cheng Y, et al. Phase III trial of linifanib versus sorafenib in patients with advanced hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2013;31(4_suppl):249.
[15] Boige V, Malka D, Bourredjem A, Dromain C, Baey C, Jacques N, et al. Efficacy, safety, and biomarkers of single-agent bevacizumab therapy in patients with advanced hepatocellular carcinoma. Oncologist 2012;17(8):1063–72.
[16] Siegel AB, Cohen EI, Ocean A, Lehrer D, Goldenberg A, Knox JJ, et al. Phase II trial evaluating the clinical and biologic effects of bevacizumab in unresectable hepatocellular carcinoma. J Clin Oncol 2008;26(18):2992–8.
[17] Zhu AX, Ryoo B, Yen C, Kudo M, Poon R, Pastorelli D, et al. Ramucirumab as second-line treatment in patients with advanced hepatocellular carcinoma following first-line therapy with sorafenib: Results from the randomized phase III REACH study. ESMO Congress 2014 2014:LBA16.
[18] Llovet JM, Decaens T, Raoul JL, Boucher E, Kudo M, Chang C, et al. Brivanib in patients with advanced hepatocellular carcinoma who were intolerant to sorafenib or for whom sorafenib failed: results from the randomized phase III BRISK-PS study. J Clin Oncol 2013;31(28):3509–16.
[19] Hsu C, Yang TS, Huo TI, Hsieh RK, Yu CW, Hwang WS, et al. Vandetanib in patients with inoperable hepatocellular carcinoma: a phase II, randomized, double-blind, placebo-controlled study. J Hepatol 2012;56(5):1097–103.
[20] Alberts SR, Reid JM, Morlan BW, Farr Jr GH, Camoriano JK, Johnson DB, et al. Gemcitabine and docetaxel for hepatocellular carcinoma: a phase II North Central Cancer Treatment Group clinical trial. Am J Clin Oncol 2012;35(5):418–23.
[21] Finn RS, Cheng A, Ikeda K, Kudo M, Tamai T, Ductus CE, et al. A multicenter open-label, phase 3 trial to compare the efficacy and safety of lenvatinib (E7080) versus sorafenib in first-line treatment of subjects with unresectable hepatocellular carcinoma. J Clin Oncol 2014. Abstract TPS4153.
[22] Llovet JM. Focal gains of VEGFA: candidate predictors of sorafenib response in hepatocellular carcinoma. Cancer Cell 2014;25(5):560–2.
[23] Horwitz E, Stein I, Andreozzi M, Nemeth J, Shoham A, Pappo O, et al. Human and mouse VEGFA-amplified hepatocellular carcinomas are highly sensitive to sorafenib treatment. Cancer Discov 2014;4(6):730–43.
[24] Qin S, Bai Y, Lim HY, Thongprasert S, Chao Y, Fan J, et al. Randomized, multicenter, open-label study of oxaliplatin plus fluorouracil/leucovorin versus doxorubicin as palliative chemotherapy in patients with advanced hepatocellular carcinoma from Asia. J Clin Oncol 2013;31(28):3501–8.
[25] Abou-Alfa GK, Johnson P, Knox JJ, Capanu M, Davidenko I, Lacava J, et al. Doxorubicin plus sorafenib vs doxorubicin alone in patients with advanced hepatocellular carcinoma: a randomized trial. JAMA 2010;304(19):2154–60.
[26] Louafi S, Boige V, Ducreux M, Bonyhay L, Mansourbakht T, de Baere T, et al. Gemcitabine plus oxaliplatin (GEMOX) in patients with advanced hepatocellular carcinoma (HCC): results of a phase II study. Cancer 2007;109(7):1384–90.
[27] Zaanan A, Williet N, Hebbar M, Dabakuyo TS, Fartoux L, Mansourbakht T, et al. Gemcitabine plus oxaliplatin in advanced hepatocellular carcinoma: a large multicenter AGEO study. J Hepatol 2013;58(1):81–8.
[28] Mir O, Coriat R, Boudou-Rouquette P, Ropert S, Durand JP, Cessot A, et al. Gemcitabine and oxaliplatin as second-line treatment in patients with hepatocellular carcinoma pre-treated with sorafenib. Med Oncol 2012;29(4):2793–9.
[29] Patrikidou A, Sinapi I, Regnault H, Fayard F, Bouattour M, Fartoux L, et al. Gemcitabine and oxaliplatin chemotherapy for advanced hepatocellular carcinoma after failure of anti-angiogenic therapies. Invest New Drugs 2014;32(5):1028–35.
[30] Assenat E, Boige V, Thezenas S, Pageaux G-P, Peron J-M, Becouarn Y, et al. Sorafenib (S) alone versus S combined with gemcitabine and oxaliplatin (GEMOX) in first-line treatment of advanced hepatocellular carcinoma (HCC): final analysis of the randomized phase II GONEXT trial (UNICANCER/FFCD PRODIGE 10 trial). ASCO Meeting Abstracts 2013;31(15_suppl):4028.
[31] Yau TC, Cheung FY, Lee F, Choo SP, Wong H, Toh HC, et al. A multicenter phase II study of sorafenib, capecitabine, and

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

oxaliplatin (SECOX) in patients with advanced hepatocellular carcinoma: final results of Hong Kong-Singapore Hepatocellular Carcinoma Research Collaborative Group study. ASCO Meeting Abstracts 2013;31(15_suppl):4117.
[32] Buckley AF, Burgart LJ, Sahai V, Kakar S. Epidermal growth factor receptor expression and gene copy number in conventional hepatocellular carcinoma. Am J Clin Pathol 2008;129(2):245–51.
[33] Blivet-Van Eggelpoel MJ, Chettouh H, Fartoux L, Aoudjehane L, Barbu V, Rey C, et al. Epidermal growth factor receptor and HER-3 restrict cell response to sorafenib in hepatocellular carcinoma cells. J Hepatol 2012;57(1):108–15.
[34] Ezzoukhry Z, Louandre C, Trecherel E, Godin C, Chauffert B, Dupont S, et al. EGFR activation is a potential determinant of primary resistance of hepatocellular carcinoma cells to sorafenib. Int J Cancer 2012;131(12):2961–9.
[35] Thomas MB, Morris JS, Chadha R, Iwasaki M, Kaur H, Lin E, et al. Phase II trial of the combination of bevacizumab and erlotinib in patients who have advanced hepatocellular carcinoma. J Clin Oncol 2009;27(6):843–50.
[36] Kaseb AO, Garrett-Mayer E, Morris JS, Xiao L, Lin E, Onicescu G, et al. Efficacy of bevacizumab plus erlotinib for advanced hepatocellular carcinoma and predictors of outcome: final results of a phase II trial. Oncology 2012;82(2):67–74.
[37] Philip PA, Mahoney MR, Holen KD, Northfelt DW, Pitot HC, Picus J, et al. Phase 2 study of bevacizumab plus erlotinib in patients with advanced hepatocellular cancer. Cancer 2012;118(9):2424–30.
[38] Yau T, Wong H, Chan P, Yao TJ, Pang R, Cheung TT, et al. Phase II study of bevacizumab and erlotinib in the treatment of advanced hepatocellular carcinoma patients with sorafenib-refractory disease. Invest New Drugs 2012;30(6):2384–90.
[39] Hsu CH, Kang YK, Yang TS, Shun CT, Shao YY, Su WC, et al. Bevacizumab with erlotinib as first-line therapy in Asian patients with advanced hepatocellular carcinoma: a multicenter phase II study. Oncology 2013;85(1):44–52.
[40] Govindarajan R, Siegel E, Makhoul I, Williamson S. Bevacizumab and erlotinib in previously untreated inoperable and metastatic hepatocellular carcinoma. Am J Clin Oncol 2013;36(3):254–7.
[41] Chiorean EG, Ramasubbaiah R, Yu M, Picus J, Bufill JA, Tong Y, et al. Phase II trial of erlotinib and docetaxel in advanced and refractory hepatocellular and biliary cancers: Hoosier Oncology Group GI06-101. Oncologist 2012;17(1):13.
[42] Asnacios A, Fartoux L, Romano O, Tesmoingt C, Louafi SS, Mansoubakht T, et al. Gemcitabine plus oxaliplatin (GEMOX) combined with cetuximab in patients with progressive advanced stage hepatocellular carcinoma: results of a multicenter phase 2 study. Cancer 2008;112(12):2733–9.
[43] O'Dwyer PJ, Giantonio BJ, Levy DE, Kauh JS, Fitzgerald DB, Benson AB. Gefitinib in advanced unresectable hepatocellular carcinoma: results from the Eastern Cooperative Oncology Group's Study E1203. ASCO Meeting Abstracts 2006;24(18_suppl):4143.
[44] Zhu AX, Stuart K, Blaszkowsky LS, Muzikansky A, Reitberg DP, Clark JW, et al. Phase 2 study of cetuximab in patients with advanced hepatocellular carcinoma. Cancer 2007;110(3):581–9.
[45] Zhu AX, Rosmorduc O, Evans J, Ross P, Santoro A, Carrilho FJ, et al. Search: a phase III, randomized, double-blind, placebo-controlled trial of sorafenib plus erlotinib in patients with hepatocellular carcinoma (HCC). Ann Oncol 2012;23(9).
[46] Hoshida Y, Toffanin S, Lachenmayer A, Villanueva A, Minguez B, Llovet JM. Molecular classification and novel targets in hepatocellular carcinoma: recent advancements. Semin Liver Dis 2010;30(1):35–51.
[47] Zhu AX, Kudo M, Assenat E, Cattan S, Kang Y-K, Lim HY, et al. EVOLVE-1: phase 3 study of everolimus for advanced HCC that progressed during or after sorafenib. ASCO Meeting Abstracts 2014;32(3_suppl):172.
[48] Sachdev JC, Javed AY, Weir AB, Korn RI, Gulla SM, Newbold RG, et al. A phase II study of temsirolimus in previously treated advanced hepatocellular carcinoma. ASCO Meeting Abstracts 2014. Abstract 4098.
[49] Chan SL, Mo F, Hui EP, Koh J, Chu C, Hui J, et al. A phase I study of temsirolimus as novel therapeutic drug for patients with unresectable hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2013;31(15_suppl):e15048.
[50] Kelley RK, Nimeiri HS, Munster PN, Vergo MT, Huang Y, Li CM, et al. Temsirolimus combined with sorafenib in hepatocellular carcinoma: a phase I dose-finding trial with pharmacokinetic and biomarker correlates. Ann Oncol 2013;24(7):1900–7.
[51] Knox JJ, Qin R, Strosberg JR, Kaubisch A, El-Khoueiry AB, Bekaii-Saab TB, et al. A phase II trial of temsirolimus (TEM) and bevacizumab (BEV) in patients with advanced hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2012;30(15_suppl):4099.
[52] Zhu AX, Kang Y-K, Rosmorduc O, Evans TR, Santoro A, Ross PJ, et al. Biomarker analyses and association with clinical outcomes in patients with advanced hepatocellular carcinoma (HCC) treated with sorafenib with or without erlotinib in the phase III SEARCH trial. J Clin Oncol 2014, 2014 ASCO annual meeting(Abstract 4028).
[53] Boix L, Rosa JL, Ventura F, Castells A, Bruix J, Rodes J, et al. C-met mRNA overexpression in human hepatocellular carcinoma. Hepatology 1994;19(1):88–91.
[54] Wang R, Ferrell LD, Faouzi S, Maher JJ, Bishop JM. Activation of the Met receptor by cell attachment induces and sustains hepatocellular carcinomas in transgenic mice. J Cell Biol 2001;153(5):1023–34.
[55] Xie B, Xing R, Chen P, Gou Y, Li S, Xiao J, et al. Down-regulation of c-Met expression inhibits human HCC cells growth and invasion by RNA interference. J Surg Res 2010;162(2):231–8.
[56] Kaposi-Novak P, Lee JS, Gomez-Quiroz L, Coulouarn C, Factor VM, Thorgeirsson SS. Met-regulated expression signature defines a subset of human hepatocellular carcinomas with poor prognosis and aggressive phenotype. J Clin Invest 2006;116(6):1582–95.
[57] Santoro A, Rimassa L, Borbath I, Daniele B, Salvagni S, Van Laethem JL, et al. Tivantinib for second-line treatment of advanced hepatocellular carcinoma: a randomised, placebo-controlled phase 2 study. Lancet Oncol 2013;14(1):55–63.
[58] Katayama R, Aoyama A, Yamori T, Qi J, Oh-hara T, Song Y, et al. Cytotoxic activity of tivantinib (ARQ 197) is not due solely to c-MET inhibition. Cancer Res 2013;73(10):3087–96.
[59] Verslype C, Cohn AL, Kelley RK, Yang T-S, Su W-C, Ramies DA, et al. Activity of cabozantinib (XL184) in hepatocellular carcinoma: results from a phase II randomized discontinuation trial (RDT). ASCO Meeting Abstracts 2012;30(15_suppl):4007.
[60] Yau TC, Sukeepaisarnjaroen W, Chao Y, Yen CJ, Lausoontornsiri W, Chen PJ, et al. A phase I/II study of foretinib, an oral multikinase inhibitor targeting MET, RON, AXL, TIE-2, and VEGFR in advanced hepatocellular carcinoma (HCC). J Clin Oncol 2012;30(15):4108.
[61] O'Neil BH, Bendell JC, Modiano MR, Machiels J-PH, Versola MJ, Hodge JP, et al. Phase I/II study of E7050 (golvantinib) in combination with sorafenib in patients (pts) with advanced hepatocellular carcinoma (HCC): phase I results. ASCO Meeting Abstracts 2013;31(4_suppl):294.
[62] Llovet JM, Pena CE, Lathia CD, Shan M, Meinhardt G, Bruix J, et al. Plasma biomarkers as predictors of outcome in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2012;18(8):2290–300.
[63] Breous E, Thimme R. Potential of immunotherapy for hepatocellular carcinoma. J Hepatol 2011;54(4):830–4.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

[64] Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. Nat Rev Cancer 2012;12(4):252–64.
[65] Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB, et al. Improved survival with ipilimumab in patients with metastatic melanoma. N Engl J Med 2010;363(8):711–23.
[66] Robert C, Thomas L, Bondarenko I, O'Day S, Weber J, Garbe C, et al. Ipilimumab plus dacarbazine for previously untreated metastatic melanoma. N Engl J Med 2011;364(26):2517–26.
[67] Hernandez-Gea V, Toffanin S, Friedman SL, Llovet JM. Role of the microenvironment in the pathogenesis and treatment of hepatocellular carcinoma. Gastroenterology 2013;144(3):512–27.
[68] Champiat S, Ferte C, Lebel-Binay S, Eggermont A, Soria JC. Exomics and immunogenics: bridging mutational load and immune checkpoints efficacy. Oncoimmunology 2014;3(1): e27817.
[69] Nault JC, Zucman-Rossi J. Genetics of hepatocellular carcinoma: the next generation. J Hepatol 2014;60(1):224–6.
[70] Sangro B, Gomez-Martin C, de la Mata M, Iñarrairaegui M, Garralda E, Barrera P, et al. A clinical trial of CTLA-4 blockade with tremelimumab in patients with hepatocellular carcinoma and chronic hepatitis C. J Hepatol 2013;59(1):81–8.
[71] Topalian SL, Hodi FS, Brahmer JR, Gettinger SN, Smith DC, McDermott DF, et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. N Engl J Med 2012; 366(26):2443–54.
[72] Hamid O, Robert C, Daud A, Hodi FS, Hwu WJ, Kefford R, et al. Safety and tumor responses with lambrolizumab (anti-PD-1) in melanoma. N Engl J Med 2013;369(2):134–44.
[73] Brahmer JR, Tykodi SS, Chow LQ, Hwu WJ, Topalian SL, Hwu P, et al. Safety and activity of anti-PD-L1 antibody in patients with advanced cancer. N Engl J Med 2012;366(26): 2455–65.
[74] Wang BJ, Bao JJ, Wang JZ, Wang Y, Jiang M, Xing MY, et al. Immunostaining of PD-1/PD-Ls in liver tissues of patients with hepatitis and hepatocellular carcinoma. World J Gastroenterol 2011;17(28):3322–9.
[75] Zeng Z, Shi F, Zhou L, Zhang MN, Chen Y, Chang XJ, et al. Upregulation of circulating PD-L1/PD-1 is associated with poor post-cryoablation prognosis in patients with HBV-related hepatocellular carcinoma. PLoS One 2011;6(9):e23621.
[76] Kuang DM, Zhao Q, Peng C, Xu J, Zhang JP, Wu C, et al. Activated monocytes in peritumoral stroma of hepatocellular carcinoma foster immune privilege and disease progression through PD-L1. J Exp Med 2009;206(6):1327–37.
[77] Sangro B, Crocenzi TS, Welling TH, Inarrairaegui M, Prieto J, Fuertes C, et al. Phase I dose escalation study of nivolumab (Anti-PD-1; BMS-936558; ONO-4538) in patients (pts) with advanced hepatocellular carcinoma (HCC) with or without chronic viral hepatitis. ASCO Meeting Abstracts 2013;31(15_suppl):TPS3111.
[78] Segal NH, Hamid O, Hwu WJ, Massard C, Butler M, Antonia S, et al. A phase I multi-arm dose-expansion study of the anti-programmed cell death-ligand-1 (PD-L1) antibody MEDI4736: preliminary data. ESMO Congress 2014 2014. Abstract 5888(Poster 1058PD).
[79] Heo J, Reid T, Ruo L, Breitbach CJ, Rose S, Bloomston M, et al. Randomized dose-finding clinical trial of oncolytic immunotherapeutic vaccinia JX-594 in liver cancer. Nat Med 2013;19(3):329–36.
[80] Segler A, Tsimberidou AM. Lenalidomide in solid tumors. Cancer Chemother Pharmacol 2012;69(6):1393–406.
[81] O'Neil BH, Goff LW, Kauh JS, Strosberg JR, Bekaii-Saab TS, Lee RM, et al. Phase II study of the mitogen-activated protein kinase 1/2 inhibitor selumetinib in patients with advanced hepatocellular carcinoma. J Clin Oncol 2011;29(17):2350–6.
[82] Choo SP, Ng QS, Chen WJJ, Tham CK, Yong W-P, Wang LZ, et al. A phase I/II study of AZD6244 in combination with sorafenib in advanced hepatocellular carcinoma. ASCO Meeting Abstracts 2012;30(15_suppl):4100.
[83] Lim HY, Yen C-J, Tak W-Y, Heo J, Choi HJ, Lin C-Y, et al. A phase II trial of MEK inhibitor BAY 86-9766 in combination with sorafenib as first-line systemic treatment for patients with unresectable hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2012;30(15_suppl):4103.
[84] Ruden M, Puri N. Novel anticancer therapeutics targeting telomerase. Cancer Treat Rev 2013;39(5):444–56.
[85] Nault JC, Calderaro J, Tommaso LD, Balabaud C, Zafrani ES, Bioulac-Sage P, et al. TERT promoter mutation is an early somatic genetic alteration in the transformation of premalignant nodules in hepatocellular carcinoma on cirrhosis. Hepatology 2014;60(6):1983–92.
[86] Turner N, Grose R. Fibroblast growth factor signalling: from development to cancer. Nat Rev Cancer 2010;10(2):116–29.
[87] Sawey ET, Chanrion M, Cai C, Wu G, Zhang J, Zender L, et al. Identification of a therapeutic strategy targeting amplified FGF19 in liver cancer by Oncogenomic screening. Cancer Cell 2011;19(3):347–58.
[88] Ahn S-M, Jang SJ, Shim JH, Kim D, Hong S-M, Sung CO, et al. Genomic portrait of resectable hepatocellular carcinomas: implications of RB1 and FGF19 aberrations for patient stratification. Hepatology 2014;60(6):1972–82.
[89] Dienstmann R, Andre F, Soria J, Tabernero J, De Braud FGM, Cereda R, et al. Significant antitumor activity of E-3810, a novel Fgfr and Vegfr inhibitor, in patients with Fgfr1 amplified breast cancer. Ann Oncol 2012;23:116–7.
[90] Abou-Alfa GK, Capanu M, O'Reilly EM, Ma J, Chou JF, Gansukh B, et al. A phase II study of cixutumumab (IMC-A12, NSC742460) in advanced hepatocellular carcinoma. J Hepatol 2014;60(2):319–24.
[91] Faivre SJ, Fartoux L, Bouattour M, Bumsel F, Dreyer C, Raymond E, et al. A phase I study of AVE1642, a human monoclonal antibody-blocking insulin-like growth factor-1 receptor (IGF-1R), given as a single agent and in combination with sorafenib as first-line therapy in patients with advanced hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2011;29(4_suppl):270.
[92] Ciuleanu TE, Bazin I, Lungulescu D, Miron L, Bondarenko I, Deptala A, et al. Final analysis: randomized, blinded, placebo-controlled phase II trial of sorafenib with and without mapatumumab in patients with advanced hepatocellular carcinoma. ASCO Meeting Abstracts 2014. Abstract 4029.
[93] Libbrecht L, Severi T, Cassiman D, VanderBorght S, Pirenne J, Nevens F, et al. Glypican-3 expression distinguishes small hepatocellular carcinomas from cirrhosis, dysplastic nodules, and focal nodular hyperplasia-like nodules. Am J Surg Pathol 2006;30(11):1405–11.
[94] Capurro M, Wanless IR, Sherman M, Deboer G, Shi W, Miyoshi E, et al. Glypican-3: a novel serum and histochemical marker for hepatocellular carcinoma. Gastroenterology 2003;125(1):89–97.
[95] Zhu AX, Gold PJ, El-Khoueiry AB, Abrams TA, Morikawa H, Ohishi N, et al. First-in-man phase I study of GC33, a novel recombinant humanized antibody against glypican-3, in patients with advanced hepatocellular carcinoma. Clin Cancer Res 2013;19(4):920–8.
[96] Yen CJ, Daniele B, Kudo M, Merle P, Park JW, Ross P, et al. Randomized phase II trial of intravenous RO5137382/GC33 at 1600 mg every other week and placebo in previously treated patients with unresectable advanced hepatocellular carcinoma. J Clin Oncol 2014, 2014 ASCO Annual Meeting(Abstract 4102).
[97] Abou-Alfa G, Yen CJ, Carrasquillo JA, Hsu C, Gansukh B, Ma J, et al. Phase Ib study of RO2137382/GC33 in combination with sorafenib in patients with advanced hepatocellular carcinoma. J Clin Oncol 2014. Abstract 4100.
[98] Iredale JP. Models of liver fibrosis: exploring the dynamic nature of inflammation and repair in a solid organ. J Clin Invest 2007;117(3):539–48.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

[99] Faivre SJ, Santoro A, Kelley RK, Merle P, Gane E, Douillard J-Y, et al. A phase 2 study of a novel transforming growth factor-beta (TGF-{beta}1) receptor I kinase inhibitor, LY2157299 monohydrate (LY), in patients with advanced hepatocellular carcinoma (HCC). ASCO Meeting Abstracts 2014;32(3_suppl): LBA173.
[100] Dillon BJ, Prieto VG, Curley SA, Ensor CM, Holtsberg FW, Bomalaski JS, et al. Incidence and distribution of argininosuccinate synthetase deficiency in human cancers: a method for identifying cancers sensitive to arginine deprivation. Cancer 2004;100(4):826–33.
[101] Cheng PN, Lam TL, Lam WM, Tsui SM, Cheng AW, Lo WH, et al. Pegylated recombinant human arginase (rhArg-peg5,000mw) inhibits the *in vitro* and *in vivo* proliferation of human hepatocellular carcinoma through arginine depletion. Cancer Res 2007;67(1):309–17.
[102] Yang TS, Lu SN, Chao Y, Sheen IS, Lin CC, Wang TE, et al. A randomised phase II study of pegylated arginine deiminase (ADI-PEG 20) in Asian advanced hepatocellular carcinoma patients. Br J Cancer 2010;103(7):954–60.
[103] Glazer ES, Piccirillo M, Albino V, Di Giacomo R, Palaia R, Mastro AA, et al. Phase II study of pegylated arginine deiminase for non-resectable and metastatic hepatocellular carcinoma. J Clin Oncol 2010;28(13):2220–6.
[104] Yau T, Cheng PN, Chan P, Chan W, Chen L, Yuen J, et al. A phase 1 dose-escalating study of pegylated recombinant human arginase 1 (Peg-rhArg1) in patients with advanced hepatocellular carcinoma. Invest New Drugs 2013;31(1):99–107.
[105] Yau TC, Cheng PN, Chan P, Pang R, Poon RT. Preliminary efficacy, safety, pharmacokinetics, and quality of life study of pegylated recombinant human arginase 1 in patients with advanced hepatocellular carcinoma. J Clin Oncol 2014. Abstract e15137.
[106] Izzo F, Marra P, Beneduce G, Castello G, Vallone P, De Rosa V, et al. Pegylated arginine deiminase treatment of patients with unresectable hepatocellular carcinoma: results from phase I/II studies. J Clin Oncol 2004;22(10):1815–22.
[107] Yeo W, Chung HC, Chan SL, Wang LZ, Lim R, Picus J, et al. Epigenetic therapy using belinostat for patients with unresectable hepatocellular carcinoma: a multicenter phase I/II study with biomarker and pharmacokinetic analysis of tumors from patients in the Mayo Phase II Consortium and the Cancer Therapeutics Research Group. J Clin Oncol 2012;30(27):3361–7.
[108] Bitzer M, Horger M, Ganten TM, Lauer UM, Woerns MA, Siveke JT, et al. Efficacy, safety, tolerability, and PK of the HDAC inhibitor resminostat in sorafenib-refractory hepatocellular carcinoma (HCC): phase II SHELTER study. ASCO Meeting Abstracts 2012;30(15_suppl):4115.
[109] Llovet JM, Hernandez-Gea V. Hepatocellular carcinoma: reasons for phase III failure and novel perspectives on trial design. Clin Cancer Res 2014;20(8):2072–9.
[110] Ding L, Ley TJ, Larson DE, Miller CA, Koboldt DC, Welch JS, et al. Clonal evolution in relapsed acute myeloid leukaemia revealed by whole-genome sequencing. Nature 2012;481(7382): 506–10.
[111] Pleasance ED, Cheetham RK, Stephens PJ, McBride DJ, Humphray SJ, Greenman CD, et al. A comprehensive catalogue of somatic mutations from a human cancer genome. Nature 2010;463(7278):191–6.
[112] Parsons DW, Jones S, Zhang X, Lin JC, Leary RJ, Angenendt P, et al. An integrated genomic analysis of human glioblastoma multiforme. Science 2008;321(5897):1807–12.
[113] Berger MF, Lawrence MS, Demichelis F, Drier Y, Cibulskis K, Sivachenko AY, et al. The genomic complexity of primary human prostate cancer. Nature 2011;470(7333):214–20.
[114] Ley TJ, Mardis ER, Ding L, Fulton B, McLellan MD, Chen K, et al. DNA sequencing of a cytogenetically normal acute myeloid leukaemia genome. Nature 2008;456(7218):66–72.
[115] Cleary SP, Jeck WR, Zhao X, Chen K, Selitsky SR, Savich GL, et al. Identification of driver genes in hepatocellular carcinoma by exome sequencing. Hepatology 2013;58(5):1693–702.
[116] Kan Z, Zheng H, Liu X, Li S, Barber TD, Gong Z, et al. Whole-genome sequencing identifies recurrent mutations in hepatocellular carcinoma. Genome Res 2013;23(9):1422–33.
[117] Guichard C, Amaddeo G, Imbeaud S, Ladeiro Y, Pelletier L, Maad IB, et al. Integrated analysis of somatic mutations and focal copy-number changes identifies key genes and pathways in hepatocellular carcinoma. Nat Genet 2012;44(6):694–8.
[118] COSMIC, <http://cancer.sanger.ac.uk/cancergenome/projects/cosmic/>.
[119] Chapman PB, Hauschild A, Robert C, Haanen JB, Ascierto P, Larkin J, et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N Engl J Med 2011;364(26):2507–16.
[120] Shaw AT, Kim DW, Nakagawa K, Seto T, Crino L, Ahn MJ, et al. Crizotinib versus chemotherapy in advanced ALK-positive lung cancer. N Engl J Med 2013;368(25):2385–94.
[121] Fong PC, Boss DS, Yap TA, Tutt A, Wu P, Mergui-Roelvink M, et al. Inhibition of poly(ADP-ribose) polymerase in tumors from BRCA mutation carriers. N Engl J Med 2009;361(2): 123–34.
[122] Sleijfer S, Bogaerts J, Siu LL. Designing transformative clinical trials in the cancer genome era. J Clin Oncol 2013;31(15): 1834–41.
[123] Verweij J, de Jonge M, Eskens F, Sleijfer S. Moving molecular targeted drug therapy towards personalized medicine: issues related to clinical trial design. Mol Oncol 2012;6(2):196–203.
[124] Hollebecque A, Massard C, Soria JC. Implementing precision medicine initiatives in the clinic: a new paradigm in drug development. Curr Opin Oncol 2014;26(3):340–6.
[125] Chiang DY, Villanueva A, Hoshida Y, Peix J, Newell P, Minguez B, et al. Focal gains of VEGFA and molecular classification of hepatocellular carcinoma. Cancer Res 2008;68(16):6779–88.
[126] Tsimberidou AM, Eggermont AM, Schilsky RL. Precision cancer medicine: the future is now, only better. Am Soc Clin Oncol Educ Book 2014:61–9.
[127] Tran B, Dancey JE, Kamel-Reid S, McPherson JD, Bedard PL, Brown AM, et al. Cancer genomics: technology, discovery, and translation. J Clin Oncol 2012;30(6):647–60.
[128] Ng IO, Guan XY, Poon RT, Fan ST, Lee JM. Determination of the molecular relationship between multiple tumour nodules in hepatocellular carcinoma differentiates multicentric origin from intrahepatic metastasis. J Pathol 2003;199(3):345–53.
[129] Engelman JA, Zejnullahu K, Mitsudomi T, Song Y, Hyland C, Park JO, et al. MET amplification leads to gefitinib resistance in lung cancer by activating ERBB3 signaling. Science 2007;316(5827):1039–43.
[130] Johannessen CM, Boehm JS, Kim SY, Thomas SR, Wardwell L, Johnson LA, et al. COT drives resistance to RAF inhibition through MAP kinase pathway reactivation. Nature 2010; 468(7326):968–72.
[131] Holohan C, Van Schaeybroeck S, Longley DB, Johnston PG. Cancer drug resistance: an evolving paradigm. Nat Rev Cancer 2013;13(10):714–26.
[132] Sequist LV, Waltman BA, Dias-Santagata D, Digumarthy S, Turke AB, Fidias P, et al. Genotypic and histological evolution of lung cancers acquiring resistance to EGFR inhibitors. Sci Transl Med 2011;3(75):75ra26.
[133] Choi YL, Soda M, Yamashita Y, Ueno T, Takashima J, Nakajima T, et al. EML4-ALK mutations in lung cancer that confer resistance to ALK inhibitors. N Engl J Med 2010;363(18):1734–9.
[134] Dawson SJ, Tsui DW, Murtaza M, Biggs H, Rueda OM, Chin SF, et al. Analysis of circulating tumor DNA to monitor metastatic breast cancer. N Engl J Med 2013;368(13):1199–209.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005

[135] Higgins MJ, Jelovac D, Barnathan E, Blair B, Slater S, Powers P, et al. Detection of tumor PIK3CA status in metastatic breast cancer using peripheral blood. Clin Cancer Res 2012;18(12): 3462–9.

[136] Murtaza M, Dawson SJ, Tsui DW, Gale D, Forshew T, Piskorz AM, et al. Non-invasive analysis of acquired resistance to cancer therapy by sequencing of plasma DNA. Nature 2013;497(7447): 108–12.

[137] Maheswaran S, Sequist LV, Nagrath S, Ulkus L, Brannigan B, Collura CV, et al. Detection of mutations in EGFR in circulating lung-cancer cells. N Engl J Med 2008;359(4): 366–77.

Please cite this article in press as: Hollebecque A. et al., Systemic treatment of advanced hepatocellular carcinoma: From disillusions to new horizons, Eur J Cancer (2014), http://dx.doi.org/10.1016/j.ejca.2014.12.005