Lenvatinib for use in combination with everolimus for the treatment of patients with advanced renal cell carcinoma following one prior anti-angiogenic therapy.

Summary

In patients with metastatic renal cell cancer options for second line therapies, following progression on anti-angiogenic agents, that demonstrate a survival advantage in clinical trials have been limited. Recently a number of agents have demonstrated efficacy in this setting. Here in we profile one such therapy, the combination of lenvatinib and everolimus, and discuss the expanded options for therapy available in this setting.

(1) Introduction

Renal cell cancer (RCC) accounts for 2-3% of all cancer diagnosed worldwide annually. In Europe alone there are over 84,000 new RCC diagnoses per year and RCC accounts for over 34,000 cancer related deaths per year. [1]

The pathogenesis of clear cell RCC, which accounts for approximately 80% of RCC, is characterised by inactivation of the von-Hippel-Lindau (VHL) gene on the short arm of chromosome 3. VHL functions as a tumor suppressor gene. The inactivation of VHL results in constitutive activation of the HIF (hypoxia-inducible factor) pathway. HIFs are transcription factors, physiologically they respond to an oxygen depleted cellular environment. HIFs promote angiogenesis. Activation of the HIF pathway in RCC is implicated in derangement of the cellular metabolism promoting anaerobic glycolysis, the pentose phosphate pathway and glutamine transport well above physiological levels. This in turn fuels the tumor microenvironment via gluconeogenesis of excess lactate. HIF, as part of its physiological role in wound healing, is active in promoting epithelial–mesenchymal transition.

HIF interacts directly with multiple oncogenic pathways, such as PI3K/AKT/mTOR, but also indirectly acts with innumerable oncogenic pathways via genome wide epigenetic changes resulting in inactivation of key tumor suppressor genes.

Epigenetic silencing of key tumor suppressor genes contributes significantly to RCC's characteristic phenotype of immune evasion.

Inactivation of VHL is thus recognised as a pivotal early step in tumorigenesis of RCC. [2]

Significant strides forward have been made in the last decade in identifying agents that are active in targeting the molecular abnormalities that underlie RCC development. That said the sheer diversity that exists in terms of molecular aberrations in RCC predisposes to the development of resistance mechanisms. This is evident in clinical practice with patients ultimately progressing on multiple lines of targeted therapy, there is as such no cure for metastatic RCC (mRCC). An unmet need exists in identifying tolerable agents that result in improvements in overall survival (OS)and ultimately in finding a cure for RCC.

A number of drugs have recently been approved for treatment of patients who have progressed on prior anti-angiogenic therapy including nivolumab, cabozantinib and the combination of everolimus and lenvatinib.

(2) Overview of the market

Prior to the development of the targeted therapies the mainstay of treatment for mRCC was interleukin 2 (IL-2). Interleukin 2's use was based on observations that it induced durable remissions in a small number of patients (10%) but at the cost of significant toxicity. Targeted therapies in mRCC broadly encompass (i) VEGF targeted tyrosine kinase inhibitors such as sunitinib, sorafenib, axitinib and pazopanib, (ii) bevacizumab a monoclonal antibody

to VEGF and (iii)agents targeting the PI3K/AKT/mTOR pathway such as everolimus and temsirolimus.[3] Recent additions to the market include the tyrosine kinase inhibitor cabozantinib and the check point inhibiter nivolumab.

A limited number of clinical trials define the setting in which each of these agents is used. Based on evidence from phase III randomised trials the European Association of urology (EAU) guidelines and the National Comprehensive cancer network (NCCN) guidelines recommend sunitinib, pazopanib or bevacizumab in combination with IFN-α be used in newly diagnosed metastatic RCC (mRCC) or advanced inoperable RCC. Temsirolimus is also an option for patients who fulfil the criteria for 'poor risk' as defined by the Memorial Sloan-Kettering Cancer Center (MSKCC) risk stratification.

The EAU guidelines and NCCN guidelines were recently updated to recognise cabozantinib and nivolumab as the new standard of care in patients who have failed VEGF targeted therapy based on two recent pivotal phase III trials. Everolimus or axitinib were previously recommended in this setting and continue to be used in clinical practice pending regulatory approval of newer agents. [4]

In the third line setting, in the absence of clinical trial options, everolimus is recommended following VEGF targeted therapy and sorafenib is recommended following mTOR targeted therapy. [3]

(2)A <u>First Line systemic treatment</u>

Sunitinib, pazopanib, temsirolimus and bevacizumab in combination with IFN-α have all demonstrated activity in the first line setting in Phase III randomised trials.

Sunitinib is a vascular endothelial growth factor receptor tyrosine kinase inhibitor. In a randomised phase III trial of 750 patients with treatment naïve mRCC sunitinib demonstrated improved median PFS compared to IFN-α, 11 months verses 5 months respectively (HR 0.42, 95% CI, 0.32 to 0.54; P<0.001). [5]At final analysis sunitinib demonstrated a numerically superior OS of 26.4 months compared to 21.8 months in the INF- α arm but did not reach statistical significance (HR 0.821; 95% CI, 0.673 to 1.001; P = .051). The failure of sunitinib to demonstrate a statistically significant improvement in overall OS may have been confounded by patient crossover to sunitinib at progression. [6]

Pazopanib is a tyrosine kinase inhibitor with targets including VEGFR, PDGFR and KIT. In a phase III, randomised, double blinded, placebo controlled trial, pazopanib demonstrated improved median PFS compared to placebo 9.2 months versus 4.2 months respectively (HR 0.46; 95% CI, 0.34 to 0.62; P < .0001). [7]The 435 patients consisted of 233 patients who were treatment naive and 202 patients who had progressed on cytokine therapy. At final analysis pazopanib was associated with an OS of 22.9 months and placebo with an OS of 20.5 months. This was not statistically significant (HR 0.91; 95% CI, 0.71-1.16; P= 0.224) but may have been confounded by the significant rate of crossover from placebo to pazopanib.[8]

In the COMPARZ trial, a phase III trial, pazopanib was shown to be non-inferior to sunitinib in terms of PFS in the first line setting, 8.4 months versus 9.5 months (HR 1.05; 95% CI 0.90 to 1.22) respectively. At final analysis OS was comparable between the arms 28.3 months for pazopanib and 29.1 months for Sunitinib (HR 0.92; 95% CI 0.79 to 1.06; P=0.24). Toxicity profiles and quality of life appeared to favour pazopanib. [9]Assessments of quality of life and toxicity similarly favoured pazopanib in the PICSES study, a double blind cross over

study. PICSES compared patient reports of preference, quality of life and toxicity in 114 patients who received pazopanib followed by sunitinib or vica versa. [10]

Bevacizumab is a monoclonal antibody to VEGF. Bevacizumab in combination with IFN- α in the first line setting has been examined in two Phase III trials. AVOREN randomised 649 patients to receive either Bevacizumab in combination with IFN- α or placebo and IFN- α. The combination of bevacizumab and INF- α resulted in a statistically significant prolongation of median PFS when compared to IFN - α, 10.4 months versus 5.4 months respectively (HR 0.63, 95% CI 0.52-0.75; p=0.0001). At final analysis median OS was 23.3 months for bevacizumab and IFN- α and 21.3 months for IFN- α and placebo. This difference in OS was not statistically significant (HR = 0.91; 95% CI, 0.76 to 1.10; *P* = .3360).[11] CALBG 90206 randomised 732 patients to receive either bevacizumab and IFN- α or IFN-α monotherapy. Median PFS in the combination arm was 8.5 months versus 5.2 months for IFN-α monotherapy (HR is 0.72 (95% CI, 0.61 to 0.83; *P* < .0001). At final analysis the addition of bevacizumab to IFN-α did not result in a statistically significant prolongation in OS. Median overall survival in the combination arm was 18.3 months and 17.4 months in the IFN-α monotherapy arm (adjusted HR 0.86, 95% CI, 0.73 to 1.01; stratified log-rank *P* = .069).[12] In both AVOREN and CALBG 90206 the authors noted that receipt of further lines of therapy following bevacizumab may have contributed to the failure to achieve a statistically significant improvement in OS.

Temsirolimus was examined in the first line setting in a phase III trial were 626 patients were randomised to receive single agent temsirolimus, single agent IFN-α or the combination of Temsirolimus and IFN-α. This trial is notable for its inclusion of patients with non-clear cell histology (20% of total study population) and its inclusion of patients with

multiple negative prognostic factors. Median overall survival in the Temsirolimus arm was 10.9 months compared to 7.3 months in the interferon arm HR 0.73 (95% CI 0.58 to 0.92; P=0.008). Median overall survival in the combination arm, 8.4 months, was inferior to single agent Temsirolimus. There was a high rate of grade 3 and grade 4 toxicity in the combination arm resulting in a substantial number of dose reductions and dose delays which may explain the inferior OS. [13]

(2)B <u>Second Line systemic treatment</u>

(2)Bi <u>Established second line systemic treatment</u>

RECORD-1, a phase III trial, randomised 416 patients with mRCC who previously progressed on sunitinib, sorafenib or both to everolimus or placebo in a 2:1 fashion. In the total population median PFS in the everolimus arm was 4.6 months versus 1.9 months in the placebo arm (HR 0.33; P < .001). In patients who had received one previous line of VEGF targeted therapy median PFS was 5.4 months for everolimus and 1.9 months for placebo (HR, 0.32; 95%confidence interval [CI], 0.24-0.43; P<.001). In patients who had received two previous lines of VEGF targeted therapy median PFS in the everolimus group was 4 months and 1,8 months in the placebo group (HR, 0.32; 95%CI, 0.19-0.54; P<.001).[14]

In the TARGET trial, a phase III randomised trial, 903 patients in the second line setting were randomised to receive either sorafenib or placebo. The majority of patients had received prior cytokine therapy. At progression patients in the placebo arm were allowed to cross over to the sorafenib arm. Median progression free survival was 5.5 months in the sorafenib group versus 2.8 months in the placebo group. At final analysis there was no difference in median OS between the two groups 17.8 months and 15.2 months for sorafenib and placebo respectively (HR = 0.88; P = .146). On censoring the patients on placebo who

crossed over to sorafenib when disease progression occurred, an OS advantage was appreciable. In this group sorafenib was associated with a median OS of 17.8 months versus 14.3 months for placebo (HR = 0.78; P = .029). [15]Sorafenib was subsequently used as the control arm for the AXIS trial.

In the AXIS trial, a randomised phase III trial, 723 patients with progression on first line Sunitinib, bevacizumab and IFN-α, temsirolimus or cytokines were randomised to receive either axitinib or sorafenib. Axitinib was shown to be superior to sorafenib in terms of PFS with axitinib having a median PFS of 6.7 months and sorafenib having a median PFS of 4.7 months (HR 0·665; 95% CI 0·544-0·812; one-sided p<0·0001). There was no statistically significant difference in median OS between axitinib and sorafenib, 15.2 months versus 16.5 months respectively (HR 0.997; CI 95 % 0.782–1.27). Subsequent lines of therapy may have contributed to this.[16]

(2)Bii <u>Recent additions to the market: Second line treatment</u>

Cabozantinib

Cabozantinib is a tyrosine kinase inhibitor which targets VEGF, MET and AXL. The METEOR trial, a phase III randomised trial, randomised 658 patients who had progressed on first line VEGF targeted therapy to receive either cabozantinib or everolimus. Cabozantinib was associated with a statistically significant improvement in median OS as compared to everolimus, 21.4 months versus 16.5 months respectively (HR 0·66 [95% CI 0·53–0·83). Cabozantinib also resulted in improved median PFS compared to everolimus, 7.4 months versus 3.9 months (HR 0·51 [95% CI 0·41–0·62]; p<0·0001).

More patients in the cabozantinib arm experienced grade 3 or grade 4 toxicity then in the everolimus arm 71% versus 60%. The most common grade 3 or above toxicities included

hypertension (15%), Diarrhoea (13%), fatigue (11%) and palmar-plantar erythrodysaesthesia (8%). Dose reductions were required in 62% of patients receiving cabozantinib and 25% of patients receiving everolimus. Treatment was discontinued for adverse events excluding disease progression in 12% of patients treated with cabozantinib and 11% of patients treated with everolimus. There was a single report of a cabozantinib related death, the cause of death was not specified.[17]

In April 2016 the FDA approved cabozantinib for patients who had received prior anti-angiogenic therapy.

In July 2016 the EMA granted market authorisation to cabozantinib for patients who progressed on prior VEGF targeted therapy.

Nivolumab

Nivolumab, an example of immune checkpoint blockade, is a fully humanised monoclonal antibody against programmed cell death 1 (PD-1). It acts to block the interaction between PD-1 and programmed cell death 1 ligands that occurs on the cell surface membrane thus blocking T cell inactivation by the cancer cell. In the Checkmate 025 study, a phase III randomised trial, 821 patients who had received one or two previous lines of anti angiogenic therapy were randomised in a 1:1 fashion to receive either Nivolumab or Everolimus. The primary end point of the study was OS. Checkmate 025 achieved its primary endpoint in that Nivolumab resulted in a statistically significant prolongation in median OS compared to everolimus, 25 months versus 19.6 months respectively 0.73 (98.5% CI, 0.57 to 0.93; P=0.002). Interestingly despite achieving an OS benefit nivolumab failed to result in a statistically significant prolongation in PFS. Forty-four percent of patients in the nivolumab arm were treated beyond progression as they were defined as deriving clinical benefit.

Nivolumab was associated with a median PFS of 4.6 months and everolimus a median PFS of 4.4 months (HR 0.88; 95% CI, 0.75 to 1.03; P=0.11). The failure to achieve a PFS benefit was not explainable by cross over at progression, only 7 patients in the everolimus arm subsequently received anti PD-1 therapy. The results for OS and PFS were numerically similar to those observed in a non-randomised Phase II trial of nivolumab. The authors noted that for patients who had not died or experienced disease progression by 6 months a subset analysis demonstrated improved PFS for nivolumab compared with everolimus.

Grade 3 or 4 toxicity occurred in 19% (n= 76) of patients who received nivolumab and 37% (n= 145) of patients who received everolimus. Grade 3 or 4 fatigue occurred in 2% of patients receiving nivolumab, the remainder of reported grade 3 or 4 toxicity occurred in ≤ 1% of the patients receiving nivolumab. Dose delays were required in 51% of patients treated with nivolumab and 66% of those treated with everolimus. Treatment was discontinued for adverse events excluding disease progression in 8% of patients treated with nivolumab and 13% of patients treated with everolimus. Treatment with nivolumab resulted in a higher proportion of patients (55%) experiencing an improved quality of life as compared to everolimus (37%). [18]

In November 2015 the FDA approved Nivolumab in mRCC in patients who had received prior anti-angiogenic treatment.

In February 2016 the EMA's Committee for Medicinal Products for Human Use (CHMP) recommended nivolumab be approved for use in patients who had progressed on prior anti-angiogenic treatment, it is currently awaiting market authorisation.

(2)C Combination therapy

The combination of lenvatinib and everolimus has been proceeded by numerous attempts to combine VEGF targeted therapies and mTOR inhibitors in the clinical trial setting. Vertical blockade of the VEGF pathway is an attractive prospect in the treatment of RCC when we consider that a number of mechanisms by which RCC develops resistance to TKIs are mediated my activation of the HIF pathway.

In a number of phase I/II trials, tyrosine kinase inhibitors such as sunitinib, sorafenib and pazopanib resulted in an unacceptable level of toxicity when combined with mTOR inhibitors such as everolimus and temsirolimus.[19-26] (TABLE 1)

The combination of bevacizumab and a mTOR inhibitor was shown to be tolerable in the phase I/II setting. [27-29]Subsequent randomised trials did not demonstrate sufficient efficacy to warrant further development of the combination.

The TORAVA trial, a French randomised phase II trial, randomised 171 patients with untreated mRCC across 3 arms: bevacizumab and temsirolimus, sunitinib, bevacizumab and interferon. Median PFS was 8.2 months (95% CI 7.0-9.6) for bevacizumab and temsirolimus, 8.2 months (95% CI 5.5-11.7) for sunitinib and 16.8 (6.0-26.0) for interferon and bevacizumab. Notably, the combination of bevacizumab and temsirolimus resulted in more toxicity then expected with 77% of patients experiencing grade 3 or above toxicity and 44% of patients experiencing serious adverse events. [30]

The INTORACT trial, a phase III randomised multicentre trial, compared the combination of Temsirolimus and bevacizumab with the combination of IFN-α and bevacizumab in 791 patients in the first line setting. INTORACT did not result in a difference in PFS between the two treatment arms. Median progression free survival in the Temsirolimus-bevacizumab arm was 9.1 months and was not superior to the median PFS in the IFN-α and Bevacizumab

arm of 9.3 months (HR 1.1; 95% CI, 0.9 to 1.3; P = .8). Median overall survival was similar in both the temsirolimus and bevacizumab arm (25.8 months) and the IFN-α and bevacizumab arm (25.5 months). [31]

RECORD-2, a phase II randomised trial, randomised 365 patients with previously untreated mRCC to receive either everolimus in combination with bevacizumab or IFN-α in combination with bevacizumab. The was no statistically significant difference in median PFS between participants treated with everolimus-bevacizumab and those treated with IFN-α and bevacizumab, 9.3 months versus 10 months respectively (HR 0.91 95% CI 0.69-1.19, p=0.485). [32]

The BEST trial, a randomised phase II trial, randomised 361 patients with mRCC to receive one of four regimens: single agent bevacizumab, Bevacizumab & Temsirolimus, bevacizumab & sorafenib or Temsirolimus & sorafenib. Patients could not have received prior anti angiogenic treatment but a single previous line of tumor vaccine or cytokine based immunotherapy was permitted. There was no statistically significant difference in median PFS between bevacizumab monotherapy (7.5 months) and any of the combination arms: bevacizumab & temsirolimus (7.6 months (HR 1.01, 95% CI 0.77-1.33, p=0.95)), bevacizumab and sorafenib (9.2 months (HR 0.89, 95% CI 0.68- 1.17, p=0.49)) temsirolimus and sorafenib (7.4 months (HR 1.07, 95% CI 0.82-1.41, p=0.68)). [33]

Subsequent generations of tyrosine kinase inhibitors, such as dovitinib [34], vatalanib[35] and tizovanib[36], were subsequently combined with mTOR inhibitors. It was speculated that unique properties of these agents may result in more tolerable combinations with clinical activity.

Dovitinib is a tyrosine kinase inhibitor that has activity in inhibiting VEGF but also fibroblast growth factor 2 (FGF-2). Fibroblast growth factor has been recognised as a possible target for overcoming resistance to VEGF targeted therapy. The combination of dovitinib and everolimus was examined in a Phase 1b trial of patients who had failed previous VEGF targeted therapy. Eighteen patients were enrolled. The MTD was dovitinib 200mg day 1-5 every 7 days and everolimus 5mg od. In total 15 people received the MTD. Two patients had a PR, 3 patients had PD as their best response, the remainder had stable disease. Median PFS was 7 months (95% CI 2.3–10 months). There was no unexpected toxicity however there was a high rate of toxicity. Overall the authors concluded that the combination had demonstrated activity but further development was limited due to the toxicity profile of the combination.[34]

Tivozanib was combined with temsirolimus in a phase 1b trial. Tivozanib targets VEGF receptor 1, VEGF receptor 2 and VEGF receptor 3. Tivozanib is known to be potent and selective. It was speculated that Tivozanib's kinase selectivity would result in less off target side effects and thus produce a tolerable combination. In total 27 patients were treated, 20 had received previous VEGF targeted therapy. No dose limiting toxicities were recorded thus the recommended phase 2 dose was temsirolimus 25mg weekly and Tivozanib 1.5 mg OD. Twenty-two patients were evaluated for response. Five patients achieved a PR and 15 patients achieved stable disease. All five patients who achieved a PR had received prior VEGF targeted therapy and 66% of those who achieved stable disease had previously received VEGF targeted therapy. The median duration of stable disease was 9.2 months. The results of this trial were promising in that both agents were combined in therapeutic doses. The combination was tolerable and had signals of possible clinical efficacy. [36] The combination of Tivozanib and everolimus warrants further investigation.

Overall numerous trials have attempted to successfully combine VEGF targeted therapy and mTOR targeted therapy. Efforts at combining therapies at therapeutic doses have been limited by toxicity. Agents which are potent and have high kinase selectivity, which result in less off target side effects, have the greatest potential for successful combination treatment in future clinical trials.

(3) Introduction to lenvatinib

(3)A Rationale for combination lenvatinib and everolimus in Renal cell cancer

Lenvatinib is a multi targeted tyrosine kinase inhibitor. In a number of preclinical studies lenvatinib has demonstrated activity in targeting pathways central to RCC neo-angiogenesis, invasion and metastases. [37]

A preclinical study involving a human breast cancer model lenvatinib was shown to be a potent inhibitor of VEGFR 3 kinase activity and VEGF- receptor 2 kinase activity. It also demonstrated inhibition of VEGFR-1, FGFR-1 and PDGFR β kinase. These receptors are active in angiogenesis and lymphangiogeneis. Dysregulation of pathways involving these receptors in malignancy promote tumor growth, invasion and metastases. [38]

Lenvatinib demonstrated activity in the KIT signaling pathway in a human small cell lung cancer model. Lenvatinib inhibited stem cell factor induced angiogenesis.[39]

Lenvatinib has demonstrated antitumor activity in RET gene fusion-driven preclinical cancer models.[40] RET expression has been noted in papillary RCC (up to 52% of patients in one study) but does not occur in clear cell RCC. The actual relevance RET expression has in papillary RCC can has yet to be determined. [41]

Everolimus inhibits mTOR a downstream effector of the PI3K/AKT/mTOR pathway which controls cell proliferation and survival. mTOR inhibitors are also known to have an immunomodulatory effect. [42, 43]

The combination of everolimus and lenvatinib targets multiple targets within two keys pathways in RCC tumorigenesis and metastases.

(3)B Chemistry

The molecular formula of lenvatinib is $C_{22}H_{23}ClN_4O_7S$.

The chemical formula of lenvatinib is 4-[3-chloro-4-(N'-cyclopropylureido)phenoxy]-7-methoxyquinoline- 6-carboxamide.

Lenvatinib has a unique binding mode with VEGFR2. Currently all approved TKI's can be classed as type 1 or type 2 inhibitors based on the conformational state of the VEGFR2 they interact with. VEGFR2 in the active state (DFG-in) forms complexes with type 1 inhibitors whereas VEGFR2 in the inactive state (DFG-out) forms complexes with type 2 inhibitors. Lenvatinib's binding mode isn't consistent with either type 1 or type 2 inhibitors and instead occupies a new distinct class of type V inhibitors. Lenvatinib binds VEGFR2 in its DFG-in status. It can bind to both the ATP binding sit and the allosteric region of VEGFR2, a characteristic that is most often related to Type 2 (DFG-out) inhibitors. These features result in lenvatinib having a prolonged residence time as compared to type 1 inhibitors but also maintaining the characteristic of kinase selectivity. The clinical implications of lenvatinib's status as a type V inhibitor have yet to be revealed but these unique characteristics may contribute to lenvatinib's potency and side effect profile as seen in clinical trials. [44]

(3)C Pharmacokinetics

In the phase I setting lenvatinib was rapidly absorbed with t_{max} (time to maximum concentration) occurring at 3 hours. C_{max} (maximum concentration) rose accordingly with escalating dose. Following multiple doses, no accumulation was observed. Following a single dose of lenvatinib measurable concentrations of lenvatinib were detectable for up to 7 days. Following a twice daily dose measurable concentrations of lenvatinib were detectable for up to 14 days. Plasma half-life was 28 hours. Elimination occurred mainly in faeces (64%) but also via urine (25%). Administration of food did not affect C_{max} but did prolong t_{max} which was 2 hours in the fasting group and 5 hours in the fed group. [45, 46]

In a phase II randomised trial Motzer et al observed that the pharmacokinetics of lenvatinib were best characterised as a three compartment model. Volume of distribution from the central compartment was 51.8 L and elimination occurred from this compartment. Volume of distribution in the two peripheral compartments was 31.2L and 43.6 L respectively. Clearance occurred at a rate of 7.4L/hour. Following oral administration of lenvatinib absorption was characterised as simultaneous first order absorption with a rate constant of 1.08/hour, a zero order absorption duration of 0.83 hours and a lag time of 0.17 hours. The observed pharmacokinetic parameters of lenvatinib were not affected by creatinine clearance or everolimus administration. [47]

In a phase 1 study in healthy patients the exposure of lenvatinib was shown to be slightly (15-19%) increased by co-administration with ketoconazole. [48]

In a three way cross over study in healthy individuals lenvatinib was not shown to cause a clinically significant prolongation in QTc interval.[49]

(4) Clinically efficacy

(4)A Phase I data Lenvatinib

In a phase 1 dose escalation study Boss et al examined the pharmacokinetics and safety of lenvatinib in 82 patients across multiple tumor types. Lenvatinib was administered once daily in a 28-day cycle with dose cohorts ranging from 0.2mg to 32mg. Maximum tolerated dose was defined as the highest dose at which one or less patients in a 6 patient cohort experiences a dose limiting toxicity. Maximum tolerated dose was reached at 25mg. At 32mg dose limiting toxicity in the form of grade 3 proteinuria occurred in 2 patients.

Of the 8 patients with RCC who received lenvatinib 4 experienced a partial response. Progression free survival for patients with RCC was 477 days (95% CI 279.0–559.0 days). [45]

(4)B <u>Phase I data Lenvatinib & Everolimus</u>

In a phase 1b study Molina et al examined the safety and maximum tolerated dose of lenvatinib combined with everolimus in 20 patients with advanced unresectable or metastatic RCC. Lenvatinib was administered at escalating doses to sequential cohorts of patients. Lenvatinib was administered at 12mg, 18mg and 24mg in combination with everolimus 5mg.Maximum tolerated dose was identified as 18mg lenvatinib. Dose limiting toxicity at the 24mg dose included nausea and vomiting and mucosal inflammation.

Seventeen patients were evaluable for tumor response, no complete responses were observed, 6 patients experienced a partial response and 10 patients experienced stable disease. This translated into a disease control rate (DCR) of 80% across all 3 cohorts and a DCR of 83.3% for patients in the maximum tolerated dose (MTD) and low dose cohorts (n=18). In the MTD and low dose cohort PFS was 330 days (95 % CI 157–446; approximately 10.9 months), PFS rate at 6 months was 72.1 % (95 % CI 48.8–95.4 % and PFS rate at 12 months was 49.5 % (95 % CI 22.7–76.2 %). [50]

(4)C <u>Phase II data Lenvatinib & Everolimus</u>

In a phase II randomised, multi-center international trial Motzer et al randomised 153 patients in a 1:1:1 fashion to receive either single agent lenvatinib (n=52), single agent everolimus (n=50) or lenvatinib and everolimus in combination (n=51).
All participants had progressed on a single previous line of VEGF targeted therapy. Participants had not received prior treatment with a PD-1 inhibitor or an mTOR inhibitor. All participants included in final analysis had metastatic disease.

The combination of lenvatinib and everolimus demonstrated a statistically significant prolongation of median PFS, the primary outcome of the study, compared to single agent everolimus, 14.6 (5·9–20·1) months versus 5.5 (3·5–7·1) months respectively (HR 0·40, 95% CI 0·24–0·68; p=0·0005). The combination arm had a numerically longer median PFS compared to the lenvatinib single agent arm 14.6 months versus 7.4 (5·6–10·2) months respectively, but this did not reach statistical significance (HR 0·66, 0·39–1·10; p=0·12). Comparison of median PFS in the two single agent arms favoured lenvatinib over everolimus (HR 0·61, 95% CI 0·38–0·98; p=0·048).

The combination of lenvatinib and everolimus achieved an objective response in 22 patients (43%), a single patient achieved a CR and 21 patients achieved a PR. This was superior to the single agent everolimus arm in which 3 patients (6%) achieved a PR [RR] 7·2, 95% CI 2·3–22·5; p<0·0001). The combination demonstrated a numerically superior objective response rate compared to single agent lenvatinib,43% versus 27% respectively, but this was not statistically significant (RR 1·6, 95% CI 0·9–2·8; p=0·10). Single agent lenvatinib was associated with a statistically significant improvement in objective response compared to single agent everolimus (RR 4·5, 95% CI 1·4–14·7; p=0·0067).

Median duration of response was 13 months in the lenvatinib and everolimus combination arm and was prolonged in comparison to both single agent everolimus (8.5 months) and single agent lenvatinib (7.5 months).

Median overall survival for the lenvatinib and everolimus combination arm was 25.5 months at primary data cut off in June 2014. This was numerically superior compared to single agent everolimus and single agent lenvatinib with median overall survivals of 18.4 months and 17.5 months respectively. This did not reach statistical significance (combination versus everolimus HR 0·55, 95% CI 0·30–1·01; p=0·062, combination versus lenvatinib HR 0·74, 95% CI 0·40–1·36; p=0·30). A post hoc updated analysis was performed in December 2014. At this time the numerical difference in median OS between lenvatinib and everolimus and single agent everolimus had reached statistical significance 25.5months versus 15.4 months respectively (HR 0·51, 95% CI 0·30–0·88; p=0·024). In this post hoc analysis there continued to be no statistically significant improvement in median OS with lenvatinib and everolimus compared to single agent lenvatinib, 25.5 months versus 19.5 months respectively (HR 0·75, 0·43–1·30; p=0·32). [47]

An ad hoc retrospective blinded independent radiological review (IRR) was performed to see if investigator assessments, and survival data based on these, were reproducible by IRR. Eighty-six events were assessed by IRR; 101 events were included in the original analysis. PFS in the combination arm was significantly prolonged compared to the everolimus arm 12.8 months versus 5.6 months respectively (HR 0·45 [95% CI 0·27–0·79]; p=0·0029). Single agent lenvatinib was associated with a numerically longer PFS compared to everolimus, 9 months versus 5.6 months, but this did not reach statistical significance (HR 0·62 [95% CI 0·37–1·04]; p=0·12). IRR assessed ORR was similar to investigator assessments. In the

combination arm 17 patients experienced a partial response and a single patient achieved a complete response. In the lenvatinib single agent arm 19 patients experienced a partial response and one patient achieved a complete response. No patients in the everolimus arm experienced a complete or partial response.[51]

(5) Safety and toxicity

In a phase I study of single agent lenvatinib common treatment related adverse events included hypertension (40%), nausea (37%), diarrhoea (34%), stomatitis (32%), proteinuria (26%), vomiting (23%) and lethargy (23%). The most commonly documented non-haematological grade 3 toxicities included hypertension (n=9, 11%) and proteinuria (n=6, 7%). Six (7%) patients experienced haematological toxicity, 3 of these were grade 3 toxicities (thrombocytopenia n=1, neutropenia n=1, febrile neutropenia n=1) and there were 2 reports of grade 4 thrombocytopenia.[45]

In a phase Ib study of lenvatinib in combination with everolimus common treatment related adverse events included fatigue (60%), mucosal inflammation (50%), diarrhoea (40%), hypertension (40%), nausea and vomiting (40%) and proteinuria (40%). Toxicity was consistent with mTOR inhibition and VEGF inhibition. There were no new safety concerns identified. Grade 3 or 4 toxicity occurred in 75% (n=15) of participants and included hypertriglyceridemia (15%), proteinuria (15%), fatigue (10%)and diarrhoea (10%). A single death occurred secondary to cholangitis in the 12mg cohort. This was not felt to be related to lenvatinib or everolimus. [50]

In a phase II randomised trial the most common grade 3 or 4 toxicities associated with single agent lenvatinib were diarrhoea (12%), hypertension (17%), proteinuria (19%), nausea (8%) and fatigue (8%). When lenvatinib was combined with everolimus common grade 3 or 4

toxicities included diarrhoea (20%), fatigue (14%), hypertension (14%), vomiting (8%), hypertriglyceridemia (8%) and anaemia (8%). Patients receiving single agent everolimus experienced the fewest grade 3 or 4 adverse events, 50% compared to 79% of those receiving single agent lenvatinib and 71% of those receiving the combination. A single treatment related death occurred in the combination arm an intracranial bleed. A myocardial infarction resulting in death in the single agent lenvatinib group was classified as being possibly related to study drug. Dose reductions were necessary in 71% of patients receiving combination therapy and 62% of those receiving single agent lenvatinib. Toxicity requiring dose reduction occurred relatively early in treatment with 49% of patients receiving combination therapy and 38% of patients receiving single agent lenvatinib undergoing a dose reduction in the first 3 cycles. Toxicity was consistent with previous earlier phase trials and there were no new safety concerns raised. [47]

(6) Regulatory Affairs

In May 2016 the FDA approved the use of lenvatinib in combination with everolimus in patients with advanced RCC who have received one prior anti-angiogenic therapy.

In July 2016 the EMA granted market authorisation to lenvatinib in combination with everolimus in patients with advanced RCC who had progressed on a previous line of VEGF targeted therapy.

(7) Expert Commentary and Five-year review

The recent additions of cabozantinib, nivolumab and combination everolimus-lenvatinib to the market will alter our current treatment algorithm for mRCC progressing on first line VEGF targeted therapy.

Cabozantinib, nivolumab and combination lenvatinib and everolimus have all demonstrated improved efficacy over everolimus such that everolimus is no longer a reasonable agent to use for most patients in the second line setting. Everolimus will likely only be considered for second line treatment in patients where mTOR inhibition would be felt to be particularity viable or where there are specific concerns regarding toxicity of VEGF targeted therapy and immunotherapy.

The place of axitinib in our treatment algorithm is less straight forward. No trials have examined the efficacy of cabozantinib, nivolumab or combination lenvatinib and everolimus compared to axitinib. Axitinib has never been directly compared to everolimus. Axitinib demonstrated improved PFS but not improved OS in the AXIS trial[52]. Axitinib is however a very active agent in a proportion of patients with a manageable toxicity profile such that, in our opinion, axitinib should continue to be included in the decision process for treating patients in the second line setting.

In the absence of head to head trials, decisions regarding the sequencing of available agents in the second line setting will be decided by the treating physician. Factors affecting this decision, on a case by case basis, could potentially include: efficacy, toxicity, cost, performance status and responses to previous therapy.

Cabozantinib is associated with a predictable toxicity profile. Toxicity associated with combination lenvatinib and everolimus was similarly predictable. Nivolumab has been shown to be tolerable in multiple trials across multiple cancer types[53]. Our understanding of the long-term consequences of immunotherapy toxicity continues to evolve. Checkmate 374, a phase 3b/4 safety trial of nivolumab in mRCC, will be crucial in informing our knowledge of the real-world profile of nivolumab NCT02596035.

Nivolumab is an intravenous treatment with associated costs in terms of delivery of intravenous therapy. Cabozantinib and combination everolimus-lenvatinib are oral treatments. The actual price of each agent will depend on the reimbursement that is established between the pharmaceutical company and the relevant regulatory authority in each country but given that lenvatinib and everolimus includes two agents this will likely result in increased expense.

In terms of efficacy both Cabozantinib and Nivolumab have demonstrated improved OS compared to everolimus in a phase III trial. Lenvatinib in combination with everolimus has done so in a phase II setting. The OS results that reached statistical significance for combination lenvatinib and everolimus compared to everolimus were achieved in a post-hoc analysis. It is also notable that a relatively small number of patients, 24 events in total, contributed to the OS calculation of combination lenvatinib and everolimus[47]. A confirmatory phase III trial would support the use of lenvatinib and everolimus in patients who have progressed on first line VEGF targeted therapy. A recent meta-analysis attempted to compare the OS associated with cabozantinib and nivolumab from two phase III trials using four Bayesian parametric survival network meta-analysis models. The authors noted that in the models utilising a time-varying HR cabozantinib was associated with improved OS in the first 5 months but following this nivolumab was associated with improved OS[54]. It is possible that this pattern reflects the maintained complete responses and maintained partial responses that are observed in a proportion of patients receiving immunotherapy. Maturing data from Checkmate 025 will better inform us regarding the rate of maintained CRs and PRs associated with nivolumab in mRCC. Given the current trend of rapid incorporation of immunotherapy into treatment algorithms, nivolumab will likely be a preferred second line agent among many physicians.

Cabozantinib, nivolumab and combination everolimus and lenvatinib, are currently being examined in patients with mRCC in the first line setting. A phase II trial of cabozantinib compared to sunitinib in the first line setting was presented at ESMO 2016. Patients, 157 in total, with poor or intermediate risk mRCC were randomised to receive either cabozantinib or sunitinib. At median follow-up of 20.8 months cabozantinib was associated with a statistically significant prolongation in PFS, 8.2 months, compared to a PFS of 5.6 months in patients receiving sunitinib (adjusted HR 0.69, 95% CI 0.48-0.98, one-sided P = 0.012)[55]. A phase III randomised trial comparing Lenvatinib and everolimus, lenvatinib-pembrolizumab and sunitinib in the first line setting is currently recruiting NCT02811861. Checkmate 214, a randomised phase III trial, will compare combination ipilimumab and nivolumab to sunitinib in the first line setting NCT02231749. ADAPTeR, a phase II study, is currently examining the use of nivolumab prior to cytoreductive therapy NCT0244686. Several trials are examining the use of nivolumab in patients who do not have metastatic disease but have high risk renal cell cancer (NCT02595918, NCT02575222).

The landscape of current trials for patients with mRCC who have progressed on prior VEGF targeted therapy shows a trend towards combining immunotherapy with either VEGF targeted therapy or novel compounds. Notable combinations currently in clinical trials include: avelumab and axitinib (NCT02684006), nivolumab and ibrutinib (NCT02899078) and pembrolizumab and bevacizumab (NCT02348008).

It is undeniable that there has been a significant evolution in the agents we have available for the treatment of mRCC. We are still significantly limited in terms of biomarkers to help inform our decision process. In melanoma LDH has been shown to be a useful biomarker for response to PD-1 therapy[56], this has not yet been validated in renal cell cancer. The

presence of CD3+ and CD8+ tumor infiltrating cells may have a role as a biomarker for response to PD-1 therapy[57]. In Checkmate 025 PD-L1 expression was not predictive of response to nivolumab. Reliable biomarkers that are reproducible in the clinical setting are urgently needed.

Overall the therapeutic landscape of systemic therapy for RCC is currently evolving and guidelines and treatment paradigms will likely undergo significant change in the coming years. The positive signal for immunotherapy in RCC in Checkmate 025 will be a significant influence on how therapies develop in the coming years as will the positive signal for combination therapies. The agents that are currently approved in the second line setting may ultimately prove efficacious in the first line setting and the next generation of trials in the second line setting trend towards combining immunotherapy with VEGF targeting agents. Utilising our evolving arsenal of targeted therapies and immunotherapies to maximum effect will rely on the development of biomarkers and the recognition of mechanisms of resistance in tailoring sequential and combination therapy in the future.

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(8) Key Points

1. Current first line treatments for mRCC include sunitinib, pazopanib, and bevacizumab in combination with interferon. Temsirolimus has demonstrated efficacy in patients with negative prognostic factors.
2. Historically second line therapies for mRCC have included everolimus and axitinib. Recent additions to the market will change our current treatment algorithm in the second line setting

3. Cabozantinib and Nivolumab demonstrated an OS advantage compared to everolimus in phase III randomised trials.
4. Combining VEGF targeted therapy and mTOR targeted therapy has been limited by significant toxicity.
5. Lenvatinib and everolimus, in targeting two keys pathways in RCC tumorigenesis, is the first combination therapy that has been efficacious which still maintaining tolerability.
6. Lenvatinib demonstrates unique pharmacodynamics properties which may contribute to the combinations success.
7. The combination of Lenvatinib and everolimus has demonstrated a predictable and manageable toxicity profile.
8. Lenvatinib enters the market at a time that two other agents have demonstrated activity in a similar setting in phase III randomised trials, cabozantinib and nivolumab. Each of these agents employs different strategies to improve upon previous targeted therapy.
9. The therapeutic landscape of systemic therapy for RCC is currently evolving and guidelines and treatment paradigms will likely undergo significant change in the coming years.
10. The positive signal for immunotherapy in RCC will be a significant influence on how therapies develop in the coming years as will the positive signal for combination therapies.

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