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TOPIC HIGHLIGHT

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Rationally designed treatment for metastatic colorectal cancer: Current drug development strategies

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Abstract

The therapeutic landscape of metastatic colorectal cancer (mCRC) has changed substantially with the emergence of new molecularly targeted agents (MTA) used as single agents or alongside standard chemotherapy. The use of these MTAs extended the overall survival of patients with mCRC to a level that current chemotherapeutics alone could not achieve. In addition, improvement in surgical techniques and ablation modalities offer cure to a limited subset of patients with mCRC and MTAs have been found to have a significant role here too, as they aid resectability. However, for the majority of patients, mCRC remains an invariably incurable disease necessitating continued courses of combined treatment modalities. During the course of these treatments, either cytotoxic or biological, cancer cells maintain their ability to acquire mitogenic mutations which render them resistant to treatment. Key challenges remain to identify appropriate subsets of patients who will most likely benefit from these new MTAs and effectively select these based on validated biomarkers. Moreover, better knowledge of the biology of colorectal cancer and the mechanisms *via* which it bypasses blockade of known signalling pathways will help us design better and more rational sequencing of these treatments, so that we can maximise the survivorship of mCRC patients. This review outlines treatment strategies for known molecular alterations with new MTAs and highlights some promising strategies.

Key words: Colorectal cancer; Molecularly targeted therapies; Drug development

Core tip: Abrogation of the mitogen-activated protein kinase pathway downstream epidermal growth factor receptor (EGFR) has emerged as a new potential targetable pathway in the treatment of metastatic colorectal cancer. Similarly, a variety of agents blocking the PI3K/Akt/mTOR pathway are underway. At the same time, a combinatorial inhibition of angiogenesis is being attempted with dual blockade of vascular-endothelial growth factor and c-mesenchymal-epithelial transition factor. Indications that HER-2 overactivation can confer resistance to treatment to MoAb against EGFR has revealed yet another potential target whereas PARP inhibitors are being tested for their ability to cause synthetic lethality in cancer cells with established defects in MMR system.

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INTRODUCTION

Colorectal cancer remains the third most common diag-

nosed cancer and the third leading cause of cancer death in the United States with 142820 estimated new cases in 2013[1].

The combinations of fluoropyrimidine with oxaliplatin (FOLFOX) or irinotecan (FOLFIRI) are all appropriate first-line treatments for metastatic colorectal cancer (mCRC) with the crossover to the alternate regimen upon progression[2].

The addition of bevacizumab, a humanized recombinant monoclonal antibody (moAb) which blocks the vascular-endothelial growth factor-A (VEGF-A or VEGF) to first line chemotherapy with either FOLFOX or XELOX (capecitabine, oxaliplatin) resulted in a better progression free survival (PFS) compared to chemotherapy alone[3]. There is also now evidence that the use of bevacizumab beyond first-line chemotherapy progression is increasing overall survival (OS) when added to second-line chemotherapy[4].

Similarly, the MoAb against the epidermal growth factor receptor (EGFR) cetuximab is approved for use in the first-line setting for KRAS wild-type (wt) mCRC combined with FOLFIRI, based on results of the CRYSTAL study[5]. Panitumumab, a fully humanised EGFR-antibody, showed superior PFS and overall survival (OS) compared with FOLFOX alone in the first-line setting[6]. The use of EGFR antibodies is restricted to patients without an activating mutation in exon 2 of *KRAS* gene (KRAS wt) which confers intrinsic resistance of cancer cells to EGFR blockade[7].

APPROVED NEW MOLECULARLY TARGETED AGENTS

Based on principles set by EGFR and VEGF inhibition, new targeted agents that abrogate multiple receptors emerged, in expectation of achieving greater results than their predecessors. And yet, their clinical effect can be characterised as modest. Questions arise as to whether these agents would exhibit their maximal effect only when used in selected patient subsets based on relevant biomarker.

Aflibercept is a recombinant fusion protein (VEGR-trap) that blocks the activity of VEGF-A, VEGF-B and placental growth factor (PlGF). In the VELOUR study, aflibercept was combined with FOLFIRI *vs* FOLFIRI/placebo in patients with mCRC patients who relapsed after treatment with oxaliplatin-containing regimen. Compared to the control group, it produced better PFS (6.9 mo *vs* 4.67 mo, HR = 0.758; $P < 0.0001$) and an overall response rate of 19.8% compared to 11.1% ($P = 0.0001$)[8].

Another new MTA is regorafenib, an inhibitor of several angiogenic and stromal receptor tyrosine kinases, including VEGFR-1, -2, -3, platelet-derived growth factor-β (PDGFR-β), fibroblast growth factor receptor 1 (FGFR-1), and TIE2. In addition, regorafenib inhibits various oncogenic receptor tyrosine kinases (RTKs: c-KIT and RET) and intracellular signaling kinases (cRAF/RAF-1, B-RAF, and B-RAF$^{V600E\ mt}$)[9]. The multicentre CORRECT phase-3 study demonstrated a 23% risk reduction of death (HR = 0.77) in mCRC patients who progressed through standard therapeutic options compared to best supportive care[10]. Undoubtedly, this study yielded a new agent for patients with no remaining therapeutic options, but clinical benefit *vs* increased toxicity has to be considered (Table 1).

NEW RATIONALLY DESIGNED TREATMENT MODALITIES

Two major pathways, the mitogen-activated protein kinase (MAPK-kinase) and the PI3K/Akt/mTOR pathways are commonly deregulated in mCRC (*i.e.*, 40% KRAS mutant, 14.5% PIK3CA mutant and 4.7% BRAF mutant[11]) and offer a good clinical rationale to target these alterations. These pathways interact with a variety of receptor tyrosine kinases (RTK), including EGFR, HER-2, hepatocyte growth factor (HGF) and others.

MAPK pathway

MAPK pathway regulates various cellular functions including cell proliferation, differentiation, migration and apoptosis. With recent successes of selected BRAF and MEK inhibitors in metastatic cutaneous melanoma these drugs have also been explored in v-Raf murine sarcoma viral oncogene homolog B1 (*BRAF*) mutant mCRC.

Vemurafenib, an oral selective inhibitor of BRAF oncoprotein was tested as single agent in a phase 1 study in patients with BRAF mutant colorectal cancer. In contrast to its pronounced activity in melanoma patients, the results of this study were comparably modest, with only 5% rate of partial responses[12]. Prahallad *et al*[13] investigated reasons for the limited single agent activity and revealed that BRAF inhibition leads to rapid upstream activation of EGFR with subsequent bypass signalling *via* the PI3K/Akt/mTOR pathway. Interestingly, preclinical BRAF mutant CRC models showed that combined BRAF/EGFR inhibition resulted in promising anti-proliferate effects and this approach is currently tested in a study, whereby vemurafenib is combined with panitumumab[14] (Table 2).

Another approach to overcome the limited response to single agent BRAF inhibitors was the addition of MEK inhibitors which resulted in synergistic anti-tumour effect in BRAF mutant CRC cell lines[15]. The rationale of this approach was that the addition of a MEK inhibitor reduces signalling *via* RAF-isoforms, especially CRAF, *via* MEK. A dual BRAF/MEK combination was clinically tested with the BRAF inhibitor, dabrafenib, in combination with the MEK inhibitor, trametinib. Despite a combined response rate of only 12% (3%CR and 9%PR), there was a significant rate of disease stabilisation (53%) with a median PFS of 3.5 mo (95%CI: 1.8-4.9)[16], justifying to test this combination in later phase trials.

Similarly to the EGFR upregulation that is observed when BRAF is inhibited, abrogation of MEK activ-

Table 1 Completed trials of approved targeted treatments

Drug	Target	Study	Phase patients	Outcome
Bevacizumab	VEGF-A	Bevacizumab in combination with Oxaliplatin-based chemotherapy as first-line therapy in metastatic CRC.	Phase Ⅲ *n* = 1401	Median OS 21.3 mo Bev/chemo arm *vs* 19.9 mo chemo alone (HR = 0.89, 97.5%CI: 0.76-1.03, *P* = 0.077)
Cetuximab	EGFR	Cetuximab combined with FOLFIRI *vs* FOLFIRI alone as first line in mCRC.	Phase Ⅲ *n* = 599	Median OS for KRAS WT patients: 23.5 mo (97.5%CI: 21.2-26.3) in Cetux/FOLFIRI arm *vs* 20 mo (97.5%CI: 17.4-21.7) in FOLFIRI alone
Panitumumab	EGFR	Panitumumab with FOLFOX *vs* FOLFOX alone in first line aCRC	Phase Ⅲ *n* = 656	Median OS (KRAS wt) 23.8 mo (97.5%CI: 20.0–27.7) with panitum/FOLFOX *vs* 19.4 mo (97.5%CI: 17.4-22.6) with FOLFOX alone
Ziv-aflibercept	VEGFA VEGFB PIGF	Addition of Aflibercept to FOLFIRI in patient with mCRC previously treated with Oxaliplatin-based chemotherapy (including patients who received Bevacizumab)	Phase Ⅲ *n* = 1226	Median OS 13.5 mo (97.5%CI: 12.5-14.9) with Aflibercept/FOLFIRI *vs* 12 mo (97.5%CI: 11-13) with FOLFIRI alone
Regorafenib	VEGFR -1, -2, -3 TIE2 KIT, RET, RAF1, BRAF, and BRAFV600E PDGFR and FGFR	Regorafenib monotherapy for previously treated metastatic colorectal cancer *vs* best supportive care (CORRECT)	Phase Ⅲ *n* = 760	Median OS 6.5 mo with Regorafenib *vs* 5 mo for best supportive care (HR = 0.77, 97.5%CI: 0.64-0.94, *P* = 0.0052)

VEGF: Vascular-endothelial growth factor; EGFR: Epidermal growth factor receptor; FGFR: Fibroblast growth factor receptor; PDGFR: Platelet-derived growth factor receptor; mCRC: Metastatic colorectal cancer; OS: Overall survival.

ity with a MEK inhibitor stimulates AKT activity in an EGFR-dependent way[17]. Therefore, a combinatorial approach is currently underway whereby the BRAF/MEK inhibitor combination of dabrafenib/trametinib is compared to dabrafenib/trametinib plus panitumumab in an ongoing phase 2 study[18] (Table 2).

Interestingly, MEK inhibitors have not displayed promising activity as single agents. However, preclinical data showed that the MEK inhibitor selumetinib (AZD6244) can augment the efficacy of conventional chemotherapeutic agents such as temozolamide or docetaxel[19]. Early results from a small phase 2 study where reported when selumetinib was combined with irinotecan in patients with KRAS and/or BRAF mutant mCRC who progressed on oxaliplatin-containing regimen. Of a total of 32 patients, there was a 10% PR rate and disease stabilisation in 52% of patients. Interestingly, the median PFS was 3.4 mo compared with the historic PFS of 2.5 mo in second line FOLFIRI[4,20].

PI3K - AKT- mTOR pathway

The PI3K-Akt-mTOR pathway deregulation is often a result of alterations in the PIK3CA gene or loss of function/expression of the tumour suppressor gene PTEN, thereby promoting cancer cell survival and motility. As reported by Perrone *et al*[21], PIK3CA/PTEN deregulation can be found in up to 27% of patients not responding to classical epidermal growth factor receptor blockade with cetuximab.

BKM120 is a pan-PI3K inhibitor with activity against all PI3K isoforms and was tested in a classical dose-escalation phase 1 study of which 43% were patients with mCRC - promisingly, 33% of the mCRC cancer patients had stable disease with good tolerability[22]. This subsequently led to the initiation of a phase-1 b study in combination with irinotecan which is currently enrolling mCRC patients[23] (Table 2).

Loss of PTEN function has been reported in up to 31% of colon cancer[24] and specific isoform inhibition of the PI3K-beta subunit has been reported to prohibit proliferation in tumours with PTEN loss[25]. As part of an ongoing phase-1 study the PI3K-beta inhibitor, GSK2636711, is currently tested in patients with advanced tumours with PTEN loss - including patients with mCRC[26] (Table 2).

The multi-tyrosine kinase inhibitor, perifosine, exhibits its main action by inhibiting signalling *via* Akt[27]. Despite promising results of perifosine in a phase 2 study in combination with capecitabine a large phase 3 study (X-PECT study) failed to show overall survival benefit in unselected mCRC patients who failed standard therapies[28]. Further trials of targeting Akt-signalling are currently underway and one of the biological rational is to combine both, Akt and MEK-inhibitors, in an attempt to horizontally target both the PI3K/Akt/mTOR and the MAP-kinase axis (Table 2).

A pharmacokinetic, biomarker-driven study using the combination of the AKT-inhibitor (MK-2206) with selumetinib (MEK-inhibitor) is currently recruiting, however early results indicate that the combination is not well tolerated mandating several dose reductions and leading to limited drug exposure in target tissue[29]. Further results will clarify whether the drug combination can enter later stage trials.

Everolimus is an oral derivative of rapamycin which inhibits the mTOR1 complex, one of the major effectors of Akt-signalling. In a phase 2 study of heavily pre-treated mCRC patients, everolimus failed to produce a meaningful PFS and OS benefit[30]. In addition a combination of everolimus and bevacizumab resulted in modest clini-

Table 2 On-going clinical trials with novel targeted therapeutics

Drug	Target	Study	Phase Patients (est.)	Objective (primary, secondary)
Vemurafenib plus Panitumumab	BRAF/EGFR	A Pilot Study of Vemurafenib and Panitumumab Combination Therapy in Patients With BRAF V600E Mutated Metastatic Colorectal Cancer	Ⅰ / Ⅱ *n* = 15	ORR PFS/OS
Vemurafenib, Cetuximab plus Irinotecan	BRAF/EGFR plus chemotherapy	A Phase Ⅰ Trial of Vemurafenib in Combination With Cetuximab and Irinotecan in Patients With BRAF V600 Mutant Advanced Solid Malignancies	Ⅰ *n* = 63	MTD
Dabrafenib, Trametinib panitumumab	BRAF/MEK and EGFR	An Open-Label, Three-Part, Phase Ⅰ / Ⅱ Study to Investigate the Safety, Pharmacokinetics, Pharmacodynamics, and Clinical Activity of the MEK Inhibitor GSK1120212, BRAF Inhibitor GSK2118436 and the Anti-EGFR antibody Panitumumab in Combination in Subjects With BRAF-mutation V600E or V600K Positive Colorectal Cancer	Ⅰ / Ⅱ *n* = 200	DLTs Pharmacokinenics RR
LGX818	BRAF	A phase Ⅰ, multicentre, open label, dose-escalation study of oral LGX818 in adult patients advanced colorectal cancer BRAF mutated	Ⅰ *n* = 126	DLT Tumour response
LGX818 BYL719 Cetuximab	BRAF PI3K EGFR	A Phase Ⅰb/ Ⅱ Multi-center, Open-label, Dose Escalation Study of LGX818 and Cetuximab or LGX818, BYL719, and Cetuximab in Patients With BRAF Mutant Metastatic Colorectal Cancer	Ⅰ / Ⅱ *n* = 124	DLT PFS
CEP-32496	BRAF	An open-Label, Phase 1/2, Single-Agent Study of CEP-32496 in Patients With Advanced Solid Tumors in Phase 1 and in Patients With Advanced Melanoma and Metastatic Colorectal Cancer With BRAF Mutation in Phase 2	Ⅰ / Ⅱ *n* = 154	ORR PFS
BKM120 Irinotecan	PI3K plus chemotherapy	Phase Ⅰ Trial of Irinotecan and BKM120 in Previously Treated Advanced Colorectal Cancer	Ⅰ *n* = 30	MTD Pharmacokinetics
BKM120	PI3K	Phase Ⅰ / Ⅱ Study of the P13Kinase Inhibitor BKM120 Given in Combination With Panitumumab in Patients With Metastatic or Advanced RAS-Wild Type Colorectal Cancer	Ⅰ / Ⅱ *n* = 40	MTD Antitumour activity
Panitumumab	EGFR			Translational research
GSK- 2636771	PI3K-beta	A Phase Ⅰ / Ⅱa, First Time in Human, Open-label Dose-escalation Study of GSK2636771 in Subjects With Advanced Solid Tumors With PTEN Deficiency	Ⅰ / Ⅱ *n* = 150	MTD Pharmacokinetics Efficacy
BKM120 MEK162	PI3K MEK	A Phase Ⅰb, Open-label, Multi-center, Dose-escalation and Expansion Study of an Orally Administered Combination of BKM120 Plus MEK162 in Adult Patients With Selected Advanced Solid Tumors	Ⅰ *n* = 88	Rate of DLTs ORR and PFS
MK-2206	Akt	A Phase 2 Study of MK-2206 in Previously Treated Metastatic Colorectal Cancer Patients Enriched for PTEN Loss and PIK3CA Mutation	Ⅱ *n* = 54	ORR Biomarker validation
MK-2206 Selumetinib	Akt MEK	Pilot Study of the Combination of MK-2206, an AKT Inhibitor, and AZD6244, a MEK Inhibitor, in Patients With Advanced Colorectal Carcinoma	Ⅱ *n* = 38	Evaluate reduction of pAKT, pERK and Ki-67
Everolimus Irinotecan Cetuximab	mTOR EGFR plus chemotherapy	Phase Ⅰ /Randomized Phase Ⅱ Study of Second Line Therapy With Irinotecan and Cetuximab With or Without RAD001, an Oral mTOR Inhibitor for Patients With Metastatic Colorectal Cancer	Ⅰ / Ⅱ *n* = 41	MTD ORR
SAR245408 (XL147) MSC 1936369B	mTOR MEK	A Phase 1 Dose Escalation Study of Combination Therapy With Oral SAR245408 (XL147) and Oral MSC1936369B in Patients With Locally Advanced or Metastatic Solid Tumors	Ⅰ *n* = 170	MTD Pharmacokinetics
Onartuzumab Bevacizumab FOLFOX	c-MET VEGF-A plus chemotherapy	Randomized, Double-Blind, Phase Ⅱ Study of FOLFOX/Bevacizumab With Onartuzumab (MetMAb) *vs* Placebo as First-Line Treatment for Patients With Metastatic Colorectal Ca	Ⅱ *n* = 196	PFS Response rate
Trastuzumab Pertuzumab Lapatinib	HER2 EGFR	A Phase Ⅱ multi-center 2-sequential cohorts trial, designed to assess the objective response rate of the anti HER2 monoclonal antibody trastuzumab, used in combination with either the small molecule tyrosine kinase inhibitor lapatinib (Cohort A) or the monoclonal antibody pertuzumab (Cohort B), in advanced disease CRC patients harbouring an amplified HER2 tumor	Ⅱ *n* = 54	ORR PFS
Olaparib	PARP	Phase Ⅱ, Open-Label, Multicenter Trial to Assess the Efficacy and Safety of the PARP Inhibitor, Olaparib, Alone in Previously-Treated Patients With Stage Ⅳ, Measurable Colorectal Cancer, Stratified by MSI Status	Ⅱ *n* = 33	Tumour response

ABT-888 Oxaliplatin Capecitabine	PARP	A Phase Ⅰ Study of ABT-888 in Combination With Oxaliplatin and Capecitabine in Advanced Solid Tumors	Ⅰ *n* = 36	MTD/DLTs Pharmacokinetics Tumour response
Brivanib Alaninate Irinotecan	VEGFR FGFR plus chemotherapy	A phase Ⅱ Study of second-line irinotecan plus brivanib, a dual tyrosine inhibitor of VEGFR and FGFR, in metastatic colorectal cancer patients enriched for elevated levels of plasma FGF following progression on bevacizumab-based treatment	Ⅱ *n* = 60	PFS

VEGF: Vascular-endothelial growth factor; EGFR: Epidermal growth factor receptor; FGFR: Fibroblast growth factor receptor; PDGFR: Platelet-derived growth factor receptor; mCRC: Metastatic colorectal cancer; ORR: Objective response rate; MTD: Maximum tolerated dose; DLTs: Dose limiting toxicities; RR: Response rate; OS: Overall survival; PFS: Progression free survival.

cal benefit and clinical programmes were stopped[31].

HGF and the receptor tyrosine kinase mesenchymal-epithelial transition factor

The receptor tyrosine kinase mesenchymal-epithelial transition factor mesenchymal-epithelial transition factor (c-MET) and its ligand HGF are currently investigated as potential active targets in mCRC. Activation of the axis through c-MET, triggers downstream effectors that promote mitogenesis and growth *via* the RAS/MAPK pathway and prevent apoptosis through activating the PI3K/AKT signalling route[32]. When colorectal tumours were examined in various stages including adenomas, carcinomas and liver metastases, overexpression of the MET oncogene was detected in more than 50% of each stage[33].

In preclinical studies, c-MET expressing cancer cell lines that were exposed to the small molecule ARQ-197 (tivantinib) showed inhibition of their proliferation, as well as induction of caspase-dependent apoptosis[34]. Tivantinib is a selective non-ATP competitive inhibitor of the c-MET receptor kinase and it was further investigated in combination with irinotecan and cetuximab in patients with KRAS wt mCRC. Although early results of a small phase 1/2 study demonstrated disease control in 8 out of 9 patients[35], a recently reported phase 2 study could not confirm these initial promising results (PFS: 8.3 mo *vs* 7.3 mo, HR = 0.85, 95%CI: 0.55-1.33, stratified log-rank *P* = 0.38)[36]. Whether the non-significant 1 mo PFS benefit will change in selected subgroups, *i.e.*, high c-met expression is currently being investigated.

Cross-talk between c-MET and VEGF pathways led to the hypothesis that combined abrogation can inhibit tumour growth. c-MET promotes tumour vasculature formation through VEGF and conversely, VEGF results in HGF production which in turn activates c-MET. Onartuzumab (MetMab), a novel "one-armed" monovalent antibody which prevents antibody-induced dimerization of c-MET was tested in combination with bevacizumab in a phase 1 study and dual-agent safety was confirmed. A randomized, phase 2, placebo-controlled study evaluating onartuzumab in combination with mFOLFOX6 plus bevacizumab *vs* mFOLFOX6/bevacizumab/placebo in patients with mCRC is underway and results are awaited[37] (Table 2).

HER-2 (ERBB2) signalling pathway

The HER (ErbB) family of receptors consists of EGFR, HER2 (ErbB-2), HER3 (ErbB-3) and HER4 (ErbB-4). They are the main transmembrane mediators for cell proliferation and survival *via* the activation of the MAP-kinase and PI3K/Akt/mTOR pathways[38]. Between these receptors there is a fine interaction upon ligand binding including homodimerisation or heterodimerisation, whereby EGFR is a preferred heterodimerisation partner of HER2.

Preclinical studies showed that aberrant activation of HER2 receptor can confer resistance of colorectal cancer cells to EGFR MoAbs, either *via* HER2 amplification or excessive production of the ligand heregulin[39]. Surgical specimens from patients who did not harbour KRAS/NRAS/BRAF/PIK3CA mutations but overexpressed the HER2 receptor, showed substantial tumour response when they incurred combined EGFR and HER2 inhibition[40].

In this context, the HERACLES phase-2 study investigates the role of either MoAbs trastuzumab/pertuzumab or trastuzumab/lapatinib in patients with HER2 amplified mCRC[41] (Table 2).

Lapatinib is an oral dual EGFR and HER-2 tyrosine kinase inhibitor which is already known to act synergistically with capecitabine in breast cancer. In a small phase-2 study of 29 patients with heavily pretreated mCRC, the combination produced a non-significant median PFS of 2.1 mo[42]. Notably, the patients in this study were not stratified by KRAS status which might have masked potential activity of lapatinib/capecitabine on KRAS wt mCRC patients.

Microsatellite instability

Microsatellite instability (MSI) produces a phenotype which characterises hereditary colorectal cancer, but can also be found sporadically in approximately 4% of patients with mCRC; it is the consequence of a defective mismatch repair system of proteins (MLH1, MSH2, MSH6 and PMS2)[43,44]. Recent studies suggested that the sporadic MSI occurs as a consequence of CpG island methylator phenotype (CIMP) in which high rates of MLH1 DNA is hypermethylated. Interestingly, in this context, there is a strong correlation with BRAF mutations[45]. The role of demethylating agents is currently under clinical investigation and it could be of interest in selected subgroups, *i.e.*, CIMP.

In addition, there is mounting evidence that MSI tumours are associated with a high rate of deficiency of homologous recombination due to mutations in coding microsatellites suggesting that synthetic lethality can be

achieved by using Poly-(ADP-ribose) Polymerase (PARP) inhibitors[46]. In preclinical models increased cytotoxicity could be achieved with the PARP inhibitor (ABT888) in MSI cell lines with mutant copies of *MRE11A* compared to wild type MSI[47]. Results of clinical trials of single agent PARP inhibitors (*e.g.*, Olaparib) or in combination with chemotherapy in MSI mCRC patients are awaited (Table 2).

FGFR plus VEGFR inhibition

Several data suggest that resistance to VEGFR inhibition can be promoted through hypoxia-mediated activation of other proangiogenic factors, independent of VEGF. Some of them are members of FGF family[48]. This can re-activate tumour revascularisation and growth despite VEGF blockade.

Dual inhibition of the fibroblast and vascular endothelial growth factors can be achieved with the oral small molecule tyrosine kinase inhibitor, brivanib alaninate. Brivanib alaninate produced promising phase-1 results in patients with advanced gastrointestinal malignancies with a median PFS in the range of 5 mo for responders. It progressed to phase-3 level combined with cetuximab *vs* cetuximab alone in KRAS-wild type patients with mCRC, but it failed to meet the end point of overall survival. The investigational arm had an OS of 8.8 mo *vs* 8.1 mo for cetuximab alone[49].

CONCLUSION

The integration of antibodies against EGFR and VEGF into clinical practice raised hopes that a new era of rationally designed treatments has started. The lessons learnt from the "one-size-fits-all" approach of EGFR-antibody therapies to selected subgroups (KRASwt) were tremendously helpful in understanding CRC tumour biology and led for example to identification of several molecular alterations in the MAP-kinase and PI3K-Akt-mTOR pathways. Moreover, further knowledge in particular on crosstalk between pathways, mechanisms of resistance and new targets for drug development have emerged.

Despite the recent progress, interpretation of current preclinical and clinical data is still limited by retrospective analyses, single centre experience, small sample size and lack of standardisation of diagnostic tools. As witnessed in this review several new MTAs in development were tested in unselected patient populations increasing the risk of failure. New trial designs focusing on biomarkers in selected subgroups will be essential not only for better understanding of the mechanisms of action, but also to make confident "go or no-go decisions" in the drug development process.

In this context a new "taxonomy-based" model for biomarker-driven drug development in mCRC has been suggested, whereby pathological and genomic classification of patient's tumour takes place upfront, before initiation of treatment. This so-called multi-"omic" profiling, testing for multiple biomarkers instead of one, could help in creating a stratification model for colorectal cancer, in a similar way as breast or lung cancer has been stratified. A defined molecular "signature", could be used for multiple therapeutic interventions or clinical trials.

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