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Update on optimal treatment for metastatic colorectal cancer from the AGITG expert meeting: ESMO Congress 2019

David K Lau [1], Matthew Burge [2,3], Amitesh Roy [4], Ian Chau [1], Daniel G Haller [5], Jeremy D Shapiro [6,7,] Marc Peeters [8], Nick Pavlakis [9,10], Christos S Karapetis [11], Niall C Tebbutt [12,13], Eva Segelov [6,14,] *Timothy J Price [15]

1) GI and Lymphoma Unit, Royal Marsden NHS Foundation Trust, London and Surrey, United Kingdom

2) Medical Oncology, Royal Brisbane Hospital, Brisbane, Australia

3) University of Queensland, Brisbane, Australia

4) Medical Oncology, Flinders Centre for Innovation in Cancer, Bedford Park, Australia

5) Abramson Cancer Center at the Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA, USA

6) Monash University, Melbourne, Australia

7) Medical Oncology, Cabrini Medical Centre, Melbourne, Australia

8) Medical Oncology, University Hospital Antwerp, Oncology, Edegem, Belgium

9) Medical Oncology, Royal North Shore Hospital, St Leonards, Australia

10) Sydney University, Camperdown, Sydney, Australia

11) Medical Oncology, Flinders Medical Centre, Bedford Park, Australia

12) Medical Oncology, Austin Health, Heidelberg, Australia

13) Dept of Surgery, University of Melbourne, Melbourne, Australia

14) Medical Oncology, Monash Medical Centre, Clayton, Australia

15) Medical Oncology, The Queen Elizabeth Hospital, Woodville, Australia

*Corresponding author - Timothy.price@sa.gov.au

Abstract

Introduction: Outcomes in metastatic colorectal cancer are improving, due to the tailoring of therapy enabled by better understanding of clinical behaviour according to molecular subtype.
Areas covered: A review of the literature and recent conference presentations was undertaken on the topic of systemic treatment of metastatic colorectal cancer. This review summarises expert discussion of the current evidence for therapies in metastatic colorectal cancer (mCRC) based on molecular subgrouping.
Expert opinion: EGFR-targeted and VEGF-targeted antibodies are now routinely incorporated into treatment strategies for mCRC. EGFR-targeted antibodies are restricted to patients with extended *RAS* wild-type profiles, with evidence that they should be further restricted to patients with left-sided tumours. Clinically distinct treatment pathways based on tumour *RAS*, *BRAF*, *HER2* and MMR status, are now clinically applicable. Evidence suggests therapy for additional subgroups will soon be defined; the most advanced being for patients with *KRAS* G12C mutation and gene *TRK* fusion defects.

Keywords: colorectal, metastatic, consensus, chemotherapy, MSI, BRAF, RAS

1. INTRODUCTION

Improvements in outcome for patients in metastatic colorectal cancer (mCRC) have been achieved with oxaliplatin and irinotecan added to fluoropyrimidine chemotherapy backbones, and subsequently with the introduction of anti-angiogenic agents, and for *RAS* wild type (WT) patients, anti-EGFR antibodies. A major change has been the progress of defining further subgroups, where alternate targeted treatment strategies have improved survival outcomes. Survival has also been extended with the availability of third-line agents, such as regorafenib and trifluridine/tipiracil, and the increasing role of integration of surgical resection, ablation and targeted radiotherapy techniques for oligometastatic disease. *RAS* and *BRAF* mutation status testing is now routinely used as a predictive factor to determine the application and timing of anti-EGFR antibodies. With increased understanding of molecular subgroups and growing availability of biological agents, there is a continued focus on the development of prognostic and predictive biomarkers to guide treatment of mCRC.

This article reviews current clinical trial evidence and addresses issues regarding management of patients with mCRC. The review and its recommendations follow a formal consensus meeting among eight Australian specialist clinicians and three international CRC experts in September 2019.

2. CONVENTIONAL CHEMOTHERAPY

2.1 Single agent chemotherapy and the sequential approach

Sequential use of single-agent cytotoxic chemotherapy may be a reasonable approach for a subgroup of patients with slowly progressing disease.[1] Several studies have demonstrated that initial single-agent fluoropyrimidine chemotherapy followed by combination chemotherapy is not inferior to upfront combination chemotherapy. Both the CAIRO and MRC FOCUS trials demonstrated that patients treated with first-line single-agent fluoropyrimidine did not have significantly different overall survival (OS) compared to patients receiving first-line combination chemotherapy.[2,3] A sequential treatment approach was further supported by the FFCD 2000-05 trial, in which upfront

combination chemotherapy was more toxic and not more effective than the sequential use of first-line 5-FU/leucovorin followed by second-line and third-line combination therapy.[4] This approach is typically considered for patients who have good performance status and are likely to be eligible for multiple lines of treatment, in whom: (i) low volume disease where there is low risk of rapid deterioration with progression, (ii) symptom control is not required, and (iii) there is no potentially resectable disease requiring rapid downsizing. Single agent chemotherapy may also be considered for patients who have multiple comorbidities or poor performance status and are unlikely to tolerate combination chemotherapy.

2.2 Doublet Chemotherapy

Doublet chemotherapy is generally the standard first-line approach for mCRC patients. Combination chemotherapy consists of a doublet chemotherapy backbone, including a fluoropyrimidine. Apart from different toxicity profiles, there is little difference in efficacy between fluoropyrimidine/oxaliplatin-based and fluoropyrimidine/irinotecan-based regimens, which are generally considered interchangeable.[2,5,6] Standard combinations are as follow: FOLFOX (5-FU and oxaliplatin), FOLFIRI (5-FU and irinotecan), and when the oral 5-FU pro-drug capecitabine is substituted for infusional 5-FU, CAPOX (capecitabine and oxaliplatin) and CAPIRI (capecitabine and irinotecan). The oral fluoropyrimidine S-1 in combination with irinotecan [7] or oxaliplatin [8] was demonstrated to be non-inferior to FOLFOX/ CAPOX with respect to progression free survival (PFS) when given in the first-line setting with bevacizumab.

The selection of a specific regimen will be influenced by factors such as patient preference, toxicity, prior adjuvant therapy and drug availability. The addition of biologic agents to monotherapy and combination chemotherapy regimens further improves outcomes (discussed below), and are generally recommended over cytotoxic chemotherapy alone in first-line treatment.[9]

2.3 Triplet Chemotherapy

The combination of three cytotoxic agents as triplet therapy (FOLFOXIRI) in first-line treatment of mCRC has been investigated in phase III trials, which report inconsistent survival results compared to doublet chemotherapy. In a GONO study, FOLFOXIRI was found to increase OS (22.6 vs 16.7 months, HR 0.70, *P*=0.032), PFS (9.8 vs 6.9 months, HR 0.63, *P*=0.0006) and ORR (60 vs 34%, p<0.0001) as well as R0 resection rates for liver metastases (15 vs 6%, *P*=0.033) compared to FOLFIRI.[10] Furthermore, in the phase III TRIBE study, FOLFOXIRI and bevacizumab was found to prolong PFS (12.1 vs 9.7 months, HR 0.75, *P*=0.006) and improve objective response rates (ORR) (65.1% vs 53.1, OR 1.64, *P*=0.006) in comparison to FOLFIRI and bevacizumab. [11] Updated median overall survival was 29.8 months in the FOLFOXIRI plus bevacizumab group compared with 25.8 months in the FOLFIRI plus bevacizumab group (HR 0.80 95% CI 0.65-0.98, *P*=0.03). [12] A similar benefit was seen in the TRIBE2 study which tested a pre-planned strategy of first line FOLFOXIRI and bevacizumab and rechallenge on progression compared to conventional sequential FOLFOX/bevacizumab and FOLFIRI/bevacizumab. As seen in the first TRIBE study, the benefit of triplet therapy in terms of ORR, PFS and OS were in favour of FOLFOXIRI, but there were significantly higher rates of grade 3 or 4 adverse events such as diarrhoea, neutropenia and febrile neutropenia. [13]

This contrasts with the results from a HORG trial, which did not demonstrate superiority of FOLFOXIRI compared with FOLFIRI, although the trial was potentially underpowered and did demonstrate trends towards improved survival and ORR with FOLFOXIRI. [14]

In the phase II VOLFI study, 99 patients with *RAS* WT mCRC were randomised to receive modified FOLFOXIRI with or without panitumumab. The study met its primary endpoint of ORR >75% and the panitumumab arm was associated with a significantly higher ORR (87 vs 61%), and higher rate of metastasectomy (33 vs 12%). However, the study did not demonstrate superior PFS (median 9.7 vs 9.7 months, *P* = 0.76) or OS (35.7 vs 29.8 months; *P* = 0.12) [15]

FOLFOXIRI is infrequently used for mCRC in some countries, in part due to uncertainty regarding survival benefit, but also due to questions regarding tolerance and of the impact on subsequent lines of therapy. However, triplet chemotherapy may be indicated in selected patients in whom maximising tumour response could lead to conversion to surgery, where biological agents are unavailable or contraindicated, or in patients with bulky, symptomatic tumours or *BRAF* mutant tumours.

3. Biological Agents in First-line Treatment

3.1 Anti-angiogenic Agents

Biological agents, either targeting the vascular endothelial growth factor (VEGF) pathway or the endothelial growth factor receptor (EGFR) pathway, should be considered standard options in first-, second- or third-line treatment of metastatic colon cancer depending on clinical and molecular characteristics.

Bevacizumab improves PFS when combined with both oxaliplatin- and irinotecan-based combination chemotherapy regimens and fluoropyrimidine monotherapy regimens in first-line treatment of mCRC.[16-19] When bevacizumab was given with irinotecan-based combination chemotherapy (IFL), compared to IFL alone there was an OS benefit [16] however this has not been consistently observed in prospectively randomised studies. Meta-analyses of the addition of bevacizumab to chemotherapy have concluded there is a small PFS and OS benefit with bevacizumab. [20,21] Table 1 summarises key randomised studies investigating bevacizumab in the first-line treatment of mCRC.

The role of bevacizumab as maintenance therapy in metastatic disease was first investigated in the MACRO study, which found no significant difference in PFS, OS or ORR between patients who continued chemotherapy and bevacizumab versus those who received single-agent maintenance bevacizumab, following 18 weeks of induction chemotherapy with CAPOX and bevacizumab.[22] The pivotal CAIRO3 study provided evidence for continuation of bevacizumab after 18 weeks of oxaliplatin-based induction chemotherapy, by demonstrating that maintenance capecitabine and

bevacizumab prolonged PFS compared to observation in patients who achieved stable disease or better after first-line treatment with CAPOX and bevacizumab.[23] Results from the PRODIGE 9 trial showed no benefit of single agent bevacizumab versus observation following 24 weeks of FOLFIRI plus bevacizumab chemotherapy. These results suggest the benefit of maintenance bevacizumab may be contingent on the continuation of concurrent chemotherapy.[24] However, the optimal maintenance strategy following induction combination chemotherapy and bevacizumab remains unclear and no study has demonstrated an overall survival benefit, suggesting the effectiveness of later lines of therapy. There is less clarity whether maintenance chemotherapy plus bevacizumab alone is inferior to chemotherapy alone or subgroups who stand to benefit from this strategy. Therefore the treatment approach should be individualised to the patient, tumour characteristics and previous toxicities.

Interestingly, while combining VEGF-targeted and EGFR-targeted antibodies with chemotherapy in the first-line setting leads to inferior outcomes than bevacizumab and chemotherapy, combination biological agents may have a role in maintenance therapy.[25] In the GERCOR DREAM/OPTIMOX3 study, the addition of the EGFR-targeted tyrosine kinase inhibitor (TKI) erlotinib to bevacizumab as maintenance therapy improved OS (24.9 vs 22.1 months, HR 0.79, *P*=0.036) at the expense of increased rates of skin rash, diarrhoea and asthenia.[26] Despite these results however, this regimen has not been adopted as a standard of care.

In clinical practice, bevacizumab is generally well-tolerated and severe toxicities are rare. Most common adverse events, such as proteinuria and hypertension, are manageable with standard approaches and usually do not require dose interruption.[27] Table 2 summarises key studies investigating maintenance bevacizumab.

Today, the role and timing of bevacizumab needs to be considered in the context of patients' goals together with physical comorbidities and molecular characteristics. Ongoing investigation to identify reliable predictive biomarkers will ideally provide guidance as to the optimal use of bevacizumab

and other VEGF-directed anti-angiogenic agents in the future. Two meta-analyses could not definitively confirm the benefit of the addition of bevacizumab to chemotherapy in *KRAS* mutant mCRC. [21,28] However, the number of patients with known *KRAS* mutations receiving chemotherapy plus bevacizumab in these pooled analyses were relatively small. No prospective study has been designed to specifically investigate the value of bevacizumab by RAS mutation status.

The only real guide to timing of bevacizumab comes from the combination of *RAS* MT status and "sidedness". In the absence of definitive randomised data, it is reasonable to add bevacizumab to chemotherapy in *KRAS* mutant and/or right sided mCRC. As discussed below, left sided *RAS* WT/*BRAF* WT (+/- Her 2 negative) patients may be best treated with a chemotherapy/anti-EGFR combination for optimal survival outcome, and bevacizumab kept in this setting for second line therapy. [29,30]

3.2 *RAS* Wild Type: EGFR-targeted Antibodies in the First-line Setting

The use of anti-EGFR with chemotherapy agents in the first-line setting has demonstrated to yield high rates of ORR, early tumour shrinkage (ETS) and depth of response in *RAS* WT tumours.[31] Anti-EGFR agents should only be used in extended *RAS* WT patients based on pivotal studies revealing *RAS* MT to be a negative predictor of effect. Infusional fluoropyrimidine based doublet chemotherapy backbones should be used based on results suggesting a lack of additional benefit when anti-EGFR antibodies are added to capecitabine or bolus 5FU based regimens.[32,33] There is currently no evidence supporting anti-EGFR therapy used in combination with fluoropyrimidines alone, or with capecitabine regimens and clinical trials testing these combinations are yet to be reported. Table 3 summarises data for anti-EGFR agents in the first-line setting.

Retrospective analyses of first line trials involving anti-EGFR antibodies revealed the predictive importance of extended or all *RAS* MT status. The predictive value of extended *RAS* analysis was first demonstrated in the PRIME trial.[34] Extended *RAS* analysis including mutational status for *KRAS* exons 2-4, *NRAS* exons 2-4 and *BRAF* codon 600 was ascertained in 90% of the study population. The

addition of panitumumab to first-line FOLFOX chemotherapy in extended-*RAS* WT patients resulted in significant improvements in PFS (10.1 vs 7.9 months, HR 0.72, *P*=0.004) and OS (26.0 vs 20.2 months, HR 0.78, *P*=0.04).[35] Importantly, the addition of panitumumab to FOLFOX in patients with *RAS* mutations was associated with poorer PFS (HR 1.31, 95% CI 1.07-1.60) and OS (HR 1.25, 95% CI 1.02-1.55). An interaction test looking for a difference in the effect of adding panitumumab between *RAS* WT patients and *RAS* mutant patients was statistically significant for PFS and OS (*P*=0.01), confirming the role of extended *RAS* status as a predictive biomarker for EGFR-targeted therapy. A subsequent meta-analysis confirmed that the patients were unlikely to benefit from anti-EGFR therapy if tumours harboured extended *RAS* mutations.[36]

A subsequent systematic review summarised several trials (COIN, PRIME, OPUS and CRYSTAL) assessing whether the addition of an anti-EGFR agent to first-line chemotherapy improves outcomes amongst patients with extended *RAS* WT tumours demonstrated that adding an anti-EGFR agent significantly improved PFS (*P*<0.0001) and ORR (*P*<0.001), with a trend towards longer OS (*P*=0.07), compared with chemotherapy alone.[37] It is thus clear that extended *RAS* mutations confers lack of benefit from EGFR-targeted treatment and may even be associated with harm. Thus extended *RAS* status testing is now standard of care in patients with mCRC being considered for anti-EGFR therapy.

The optimal strategy for maintenance anti-EGFR therapy remains unclear. The phase II MACRO2 study randomised 193 patients with *KRAS* WT tumours to receive FOLFOX plus cetuximab for 4 months followed by continued therapy or cetuximab monotherapy. No statistically difference in the primary endpoint was observed (PFS at 9 months: 60% vs 72% respectively; *P*=0.0502). [38] By contrast the phase II VALENTINO study concluded de-escalation to single agent panitumumab maintenance was inferior to 5-FU plus panitumumab after 4 months of induction FOLFOX plus panitumumab. Enrolling 229 patients, the 10 month PFS was inferior in the panitumumab arm (49.0 vs 59.9%, HR 1.51 95% CI 1.11-2.07; *P*=0.01). [39] The continuation of anti-EGFR agents as first line therapy may therefore be based upon toxicity and patient goals.

3.3 *RAS* Wild Type: First-line VEGF versus EGFR antibodies in an all *RAS* population

Several studies have sought to define the optimal biologic agent for use in first-line treatment in patients with all *RAS* WT tumours. Despite inconclusive evidence overall, there is increasing evidence to suggest that primary tumour location in the left versus right colon is important when selecting a first-line biological agent.

Left versus Right Sided primary tumours

There is historic data that right- and left-sided colon cancers are different entities, with distinct clinical, demographic and histological features. Right-sided cancers, defined as those proximal to the splenic flexure, tend to occur in female and older patients, and are associated with poorly differentiated and locally advanced tumours, peritoneal carcinomatosis and worse prognosis.[40,41] There is also strong evidence that side impacts on outcomes for anti-EGFR therapy. For example the NCIC CO.17 trial, which was designed to compared cetuximab versus best supportive care in chemotherapy-refractory mCRC and found that among *KRAS* WT patients, patients with left-sided tumours had significantly improved PFS when treated with cetuximab (5.4 vs 1.8 months, HR 0.28, 95% CI 0.18-0.45, *P*<0.0001) whereas those with right-sided tumours did not (1.9 vs 1.9 months, HR 0.73, 95% CI 0.42-1.27, *P*=0.26). [41] Similar findings in later-line treatment were found in a pooled analysis of four randomised panitumumab studies. [42]

A recent meta-analysis of first-line clinical trials in mCRC confirmed that primary tumour location was predictive of survival benefit from the addition of an anti-EGFR antibody to standard chemotherapy in *RAS* WT tumours (left-sided OS HR 0.69, *P*<0.0001; right-sided OS HR 0.96, *P*=0.802).[43] With respect to choice of first-line biologic agent, patients with *RAS* WT left-sided cancers had a significant benefit from anti-EGFR treatment compared to anti-VEGF treatment (HR 0.71, 95% CI 0.58-0.85, *P*=0.0003) while patients with right-sided cancers had a non-significant improvement in OS with bevacizumab-based treatment (HR 1.3, 95% CI 0.97-1.74, 0.081). A pooled

analysis of six randomised trials (CRYSTAL, FIRE-3, CALGB 80405, PRIME, PEAK, 20050181) investigated the prognostic and predictive effects of tumours side in patients with *RAS* WT mCRC receiving first- or second-line chemotherapy with EGFR-targeted antibodies.[44] This meta-analysis not only reinforced the poorer prognosis associated with right-sided tumours, but also confirmed that anti-EGFR treatment prolonged OS (HR 0.75, 95% CI 0.67-0.84, *P*<0.001) and PFS (HR 0.78, 95% CI 0.70-0.87, *P*<0.001) and improved ORR (OR 2.12, 95% CI 1.77-2.55) in left-sided tumours only. The test for interaction between primary tumour location and EGFR-targeted antibodies was significant for OS (p<0.001) and PFS (*P*=0.002) and trended towards significance for ORR (*P*=0.07). Head-to-head comparison data of bevacizumab versus anti-EGFR antibodies in first-line mCRC are summarised in Table 4.

In line with these findings, mCRC guidelines published by the National Comprehensive Cancer Network (NCCN) in the United States of America and the National Health and Medical Research Council (NHMRC) of Australia recommend that anti-EGFR agents should only be used routinely in left-sided *RAS* WT tumours.[45,46] For potentially resectable *RAS* WT patients where conversion to resectability is the goal, the latest European Society for Medical Oncology (ESMO) guidelines recommend intensified treatment with cytotoxic doublet plus an anti-EGFR antibody to maximise R0 resection, however consideration should be made with respect to primary tumour sidedness.[47] Until the predictive value of primary tumour location is validated in prospective studies, the question of whether EGFR-targeted therapies should be offered to patients with *RAS* WT right-sided tumours is likely to remain controversial.

In addition to considerations regarding efficacy, the choice of biological agents may also be influenced by potential toxicity and logistical factors. The EGFR- and VEGF-targeted agents have different side effect profiles. Whilst the emergence of acneiform rash with anti-EGFR therapy may be associated with better outcomes [48] many patients find this toxicity severe and distressing. Furthermore, due to time and tissue requirements for *RAS* mutation testing, chemotherapy

treatment may need to commence before determining eligibility for anti-EGFR therapy. This could be mitigated by starting the chemotherapy backbone while awaiting the results of *RAS* testing.

3.4 *RAS* Wild Type: Combinations of Bevacizumab with an EGFR-targeted agent and chemotherapy

Despite promising preclinical and early phase II trials, studies testing the addition of anti-EGFR monoclonal antibodies to chemotherapy and bevacizumab in the first-line setting did not demonstrate any advantages with two biological agents.[25,49] Therefore, the use of combinations of EGFR-targeted and VEGF-targeted in mCRC is not recommended based on current evidence. Hypothesised explanations for the observed detrimental effect of combining two targeted agents include direct negative interactions between antibodies, decreased dose intensity of chemotherapy, as well as inclusion of patients with *NRAS* and *KRAS* exon 3 and 4 mutated tumours in the studies. It is unclear whether this negative interaction persists with sequential biologic therapy and whether it can impact on the efficacy of subsequent therapy.

4. Second-line and Subsequent Treatment

Decision-making when patients progress following initial treatment is becoming more complex, partly due to the blurring of traditional 'line-based' treatment approach where patients were treated until progression and then switched to salvage therapies.

4.1 Second-line Chemotherapy Strategy

Most patients who progress following a first-line oxaliplatin doublet will switch to irinotecan-based therapy, and vice versa. When chemotherapy is used alone, similar OS outcomes are produced irrespective of whether oxaliplatin-based or irinotecan-based chemotherapy is sequenced first, although responses are less frequent and PFS is shorter with second-line therapy with both approaches.[5]

4.2 Second-line Biologic Strategy

The choice of biological agent in the second-line setting is influenced by various factors, including biologic agent use in the first-line, *RAS* status and increasingly other molecular characteristics such as *BRAF* and HER2 status, as described below. Randomised trials of EGFR-targeted antibody panitumumab in the second- and third-line setting have reported outcomes by extended *RAS* mutational status among *KRAS* exon 2 WT patients. Similar to results from first-line studies described above, patients with *RAS* mutations have similar or inferior outcomes when treated with panitumumab compared to FOLFIRI alone, irinotecan alone or best supportive care. [29,50,51] Tables 3 and 5 summarise current data for second-line use of anti-EGFR and anti-VEGF agents, respectively.

Evidence from the bevacizumab, aflibercept and ramucirumab phase III second line studies support the use of 2nd line anti-angiogenic therapy after even after progression on a first-line bevacizumab containing regimen [52-54]. In the TML study, 820 patients who had previously progressed after a bevacizumab containing first line regimen (irinotecan or oxaliplatin based) were randomised to bevacizumab plus standard chemotherapy or standard chemotherapy alone. Median overall survival was 11.2 months in the bevacizumab plus chemotherapy arm and 9.8 months in chemotherapy alone (HR 0.81 95% CI 0.69-0.94, *P*=0.0062). [54] Similarly, in patients who had previously received FOLFOX/bevacizumab, the addition of the VEGFR-2 targeted antibody, ramucirumab to FOLFIRI chemotherapy prolonged overall survival (Median OS 13.3 vs 11.7 months, HR 0.844 95% CI 0.730-0.976, *P*=0.0219). [52] In the VELOUR trial, the addition of aflibercept (recombinant protein VEGF trap) to FOLFIRI chemotherapy was compared to FOLFIRI plus placebo. The study population was predominantly anti-angiogenic naïve with 30% receiving prior bevacizumab. The median overall survival was 13.50 versus 12.06 months respectively (HR 0.817 95.34% CI 0.713-0.937, *P*=0.0032). [53]

There is a lack of prospective data to suggest continuing anti-EGFR therapy in the second line setting is of significant benefit. In a phase II trial, 153 patients with *KRAS* exon 2 WT mCRC were randomised to FOLFOX plus cetuximab or FOLFOX alone after previously receiving 1st line FOLFIRI plus cetuximab. Whist the PFS was significantly longer in the FOLFOX plus cetuximab arm (median PFS 6.9 months vs 5.3 months, *P*=0.025), the OS did not reach statistical significance (median OS 23.7 vs 19.8 months, *P*=0.056). [55] For left sided *RAS* WT tumours, initially treated with anti-EGFR therapy with chemotherapy, we would recommend the addition of bevacizumab to second line chemotherapy if the patient is suitable.

In patients with *RAS* WT mCRC who have received first line bevacizumab, there is a paucity of randomised data supporting the use of an anti-EGFR agent in second line chemotherapy. In the randomised phase II PRODIGE-18 study there was a trend in favour of maintaining bevacizumab rather than switching to cetuximab following progression on a bevacizumab containing regimen (median OS 15.8 months vs 10.4 months, HR 0.69 95% CI 0.46-1.04, *P*=0.08). [56] Similarly, the SPIRITT trial which randomised 188 patients to FOLFIRI plus panitumumab or bevacizumab after progression of oxaliplatin-based chemotherapy with bevacizumab did not show a significantly significant difference between the two biologic strategies.[57] Nevertheless, it is reasonable to switch to an anti-EGFR containing regimen after failure of bevacizumab in first line.

4.3 Beyond Second-line Strategy

As anti-EGFR agents cetuximab and panitumumab are associated with clear survival advantages in the chemotherapy refractory setting [58,59], their use should be standard of care in *RAS* WT patients who have not previously been exposed to these agents, and there is no apparent clinical difference between the two anti-EGFR agents. [60]

Regorafenib is an oral multi-tyrosine kinase inhibitor that was compared with placebo in salvage setting in two randomised trials, CORRECT.[61] Treatment with regorafenib resulted in a statistically

significant but clinically modest improvement in OS (Median 6.4 vs 5.0 months HR 0.77, 95% CI 0.64-0.94, *P*=0.0052 and HR 0.55, *P*<0.0001 respectively). The median PFS was 1·9 months in the regorafenib group and 1·7 months in the placebo group (HR 0·49, 95% CI 0·42–0·58, *P*<0·0001). The main toxicities requiring dose modifications and delays were diarrhoea and asthenia, but quality of life in both study arms were similar. Similar results were replicated in an Asian cohort in the phase III CONCUR trial. [62] Evidence from two subsequent trials suggest, a dose escalation strategy where regorafenib is commenced at a lower dose with planned escalation to full dose (160mg per day) on the basis of toxicity is better tolerated without detriment to treatment efficacy. [63,64]

The RECOURSE trial investigated trifluridine-tipiracil, a synthetically engineered fluoropyrimidine, in patients who had received at least 2 prior lines of chemotherapy including fluoropyrimidine, oxaliplatin, irinotecan bevacizumab and anti-EGFR therapy, trifluridine-tipiracil prolonged OS from 5.3 for placebo to 7.1 months (HR 0.68, 95% CI 0.58-0.81, *P*<0.001) and PFS from 1.7 to 2.0 months (HR 0.48, 95% CI 0.41-0.57, *P*<0.001) There was also a delay in time to deterioration in performance status from 4.0 to 5.7 months (HR 0.66, 95% CI 0.56-0.78, *P*<0.001).[65] The most common toxicities were haematological, including ≥grade 3 neutropenia in 38% and febrile neutropenia in 4%. A post-hoc analysis of the RECOURSE study suggest the onset of neutropenia may be a positive predictor for efficacy [66].

Novel approaches to treatment sequencing are being investigated to maximise the clinical utility of existing agents including rechallenge with anti-EGFR agents. An ORR of 54% was reported when irinotecan-refractory patients who had clinical benefit with a line of cetuximab- plus irinotecan-based therapy, followed by a different chemotherapy and progression, were retreated with the same cetuximab- plus irinotecan-based therapy.[67]

5.Novel Subgroups

5.1 BRAF Mutation

The serine/threonine-protein kinase BRAF is a component of the RAS/RAF/MEK/MAP signal transduction pathway, which mediates signals from cell surface receptors to the nucleus to regular cell growth, differentiation and survival. BRAF is downstream of RAS, and is responsible for phosphorylation and activation of MEK1 and MEK2.[68] Mutations in *BRAF*, which are detected in approximately 10% of CRC patients overall, are mutually exclusive of *KRAS* mutations and occur more frequently in patients with mismatch repair deficiency (dMMR).[69-72] The vast majority of *BRAF* mutations are V600E, which results in an amino acid change from valine (V) to glutamic acid (E), leading to constitutive activation of BRAF by mimicking tyrosine kinase phosphorylation.[68] *BRAF* mutations in CRC are associated with female gender, older age and right-sided tumours with more advanced TNM stage, poor differentiation, mucinous histology, and dMMR.[73,74] *BRAF* mutated CRC also tends to metastasise more commonly to the peritoneal cavity and lymph nodes, which in one series occurred in approximately 50% of *BRAF* mutant patients compared to 25-35% of *BRAF* WT patients.[69] The clinical utility of knowing patients' *BRAF* mutation status is now accepted among oncologists, with clinical practice guidelines recommending that *BRAF* mutational status be ascertained in mCRC patients for prognostic stratification.[75] Mutant *BRAF* is also an emerging molecular target in mCRC.

BRAF V600E mutated colorectal cancers are associated with poorer prognosis, with median survival approximately half that of *BRAF* WT tumours (10-15 months).[74,76] The poorer survival associated with *BRAF* mutations may be related to lower response rates to first-line chemotherapy, but is also likely driven by subsequent rapid progression that precludes many patients from receiving second-line therapies. [77] There is also evidence that patients with *BRAF* mutations rarely have patterns of metastatic spread that are amenable to surgical resection with curative intent, and those that do undergo resection have a higher rate of subsequent relapse compared to *BRAF* WT patients.[78]

Despite the evidence for poorer prognosis associated with the most prevalent *BRAF* mutation V600E, there are clearly some patients that fare better and are able to receive second and subsequent lines of therapy. The prognosis amongst patients with non-V600 *BRAF* mutations is variable. Whilst, mutations in codons 597/601 portend a poor prognosis similar to V600E, mutations involving codons 594/596 have a prognosis similar to *BRAF* WT. [79] Preclinical evidence suggest non-V600 *BRAF* mutations are sensitive to anti-EGFR therapy [80] and requires further clinical investigation.

BRAF mutation status does not appear to be predictive of the ORR and PFS benefit afforded by the addition of bevacizumab to first-line chemotherapy in mCRC patients.[81] The utility of *BRAF* mutation status as a predictive marker of benefit from anti-EGFR directed therapy is more controversial. There have been no randomised studies designed primarily to address this issue prospectively, but retrospective analyses of randomised trials have attempted to investigate the effect of anti-EGFR antibodies in *BRAF* mutant patients. The predictive role of *BRAF* mutation status in *RAS* WT patients receiving anti-EGFR therapy has been examined in two meta-analyses, neither demonstrating a statistically significant difference in PFS or OS, with aggregate HRs approximating 0.9.[82,83] In the meta-analyses of Rowland et al, an interaction test did not demonstrate a significant effect of *BRAF* mutation status on the survival benefit associated with EGFR targeted therapies.[82] *BRAF* mutant patients receiving anti-EGFR antibody monotherapy in later lines of treatment rarely have a RECIST-defined tumour response.[84] However, the fact that *BRAF* mutations predominantly occur in right-sided tumours substantially confounds the analysis. Given recent data (discussed below), perhaps the best use of EGFR antibodies in BRAF V600E mutated patients is in combination with BRAF +/-MEK inhibitors.

Post-hoc analyses of several trials have suggested the benefit of anti-angiogenic therapy in *BRAF* mutant mCRC, however patient numbers are small. An analysis of the initial study showing bevacizumab benefit [85], showed an OS of 16 months in patients receiving chemotherapy plus bevacizumab versus 8 months for chemotherapy alone (n=10). [86] Maintenance bevacizumab with

capecitabine may be of value in comparison to observation from an analysis of the CAIRO3 trial (n= 30, Median PFS 13.0 months vs 5.7 months, HR 0.28 95% CI 0.12–0.64). [87] Post-hoc analyses of the RAISE [88] and VELOUR [89] studies suggest some benefit of continuing anti-angiogenic therapy in the 2nd line setting with ramucirumab and aflibercept respectively.

Given that *BRAF* V600E mutations are associated with poor prognosis, and lower chance of proceeding to subsequent treatment lines, it has been suggested that chemotherapy intensification with FOLFOXIRI +/- bevacizumab may be the chemotherapy of choice. This is based on a pre-planned subgroup analysis of the TRIBE study, which investigated a 'triplet' first-line approach with the addition of oxaliplatin to FOLFIRI/bevacizumab, where patients with *BRAF* mutant tumours appeared to benefit from intensified treatment (n=28, median OS: 19 vs 10.7 months, HR 0.54, 95% CI 0.24-1.20).[12] By contrast, a subgroup analysis (n=14) of the phase II randomised VOLFI study suggests *BRAF* mutant tumours may benefit from the addition of panitumumab to FOLFOXIRI chemotherapy. Response rates were 71% versus 22% in comparison to FOLFOXIRI alone, however there was no difference in median PFS (6.5 versus 6.1 months). [15] Similarly, numerically higher response rates were observed with FOLFIRI plus cetuximab in comparison to FOLFIRI plus bevacizumab (52% vs 40%) within a subgroup analysis of the FIRE-3 trial however PFS and OS were similar. [90]

In stark contrast to melanoma, single agent small molecule BRAF inhibitors at standard doses have almost no activity in mCRC.[91] The mechanism of resistance is postulated to feedback reactivation of the EGFR receptor, resulting in ongoing stimulation of the MAPK pathway.[92] This led to a focus on combination approaches, inhibiting the MAPK pathway at multiple levels, as well as parallel pathways and/or the EGFR receptor itself. Evidence of safety and activity has been demonstrated in early phase trials of combinations of BRAF with MEK inhibitors, EGFR inhibitors, PI3K inhibitors and/or chemotherapy.[93-97]

On the basis of ORR of 35% observed in a phase 1b trial of the combination of vemurafenib [98], irinotecan and cetuximab, the phase 2 SWOG 1406 study randomised 106 pre-treated mCRC

patients with extended *RAS* WT and *BRAF* V600E mutation to receive cetuximab and irinotecan (IC), with or without vemurafenib. [99] The addition of vemurafenib was associated with longer PFS (4.3 vs 2.0 months, HR 0.48, 95% CI 0.31-0.75, p=0.001), higher disease control rate (67 vs 22%, p<0.001) and a trend to improved OS (9.6 vs 5.9 months, HR 0.73, 95% CI 0.45-1.17, *P*=0.19), notwithstanding the fact that 48% of patients in the IC arm crossed over to the vemurafenib arm on progression. Interestingly, the addition of vemurafenib was associated with benefit in patients crossing over to the vemurafenib arm after progression on IC alone, with median PFS of 5.8 months and OS of 12.1 months. SWOG 1406 provides evidence supporting the use of BRAF inhibitor vemurafenib in combination with irinotecan and cetuximab in 2nd or later line treatment of mCRC with *BRAF* V600E mutations.

In a multi-cohort study, 142 patients were allocated to combined BRAF and EGFR with dabrafenib and panitumumab (D+P), triplet therapy of BRAF, MEK and EGFR inhibition with dabrafenib, trametinib and panitumumab (D+T+P) or the combination of MEK and EGFR inhibition (T+P). The ORR was 10%, 21% and 0% and median PFS was 3.5, 4.2 and 2.6 months for the D+P, D+T+P and T+P arms respectively [97]. Despite these results, development of the triplet combination was abandoned due to high toxicity and cost.

The recently reported BEACON CRC study randomised 665 patients with *BRAF* V600E mCRC who had progressed after one or two lines of chemotherapy. Patients were allocated to receive triplet (cetuximab, encorafenib, and binimetinib), doublet (cetuximab and encorafenib) or chemotherapy (cetuximab/irinotecan or FOLFIRI). The median overall survival was 9.0 months in the triplet group, compared to 5.4 months in the control chemotherapy group (HR 0.52; 95% CI 0.39-0.70; *P*<0.001). The median overall survival in the doublet group was 8.4 months (HR 0.60; 95% CI 0.45-0.79; *P*<0.001). The ORR for triplet, doublet and chemotherapy were 26, 22 and 2% respectively. [96] These results support the use of combination cetuximab, encorafenib and binimetinib in metastatic *BRAF* V600E colorectal cancer after failure of first line chemotherapy. However, given the small

differences in survival outcome between the triplet and doublet arms the value of the MEK inhibitor binimetinib remains in question. Furthermore, the optimal timing of this regimen has not been determined and the phase II ANCHOR study testing this combination in the first line setting will inform treatment sequencing (NCT03693170). Concern has been raised about the cost of these combinations and the funding mechanism for what is an uncommon clinical variant.

Based upon the body of evidence, the utility of anti-EGFR therapy in combination with chemotherapy in *BRAF* V600E mutant mCRC remains controversial. Whilst chemotherapy and bevacizumab is recommended as first line therapy in NCCN and ESMO guidelines [45,47], chemotherapy in combination with anti-EGFR therapy may also be an effective option where the therapeutic goal is cytoreduction. Anti-EGFR therapy is effective in combination with a BRAF mutant inhibitor in later lines of therapy where available.

5.2 Defective Mismatch Repair

Microsatellite instability (MSI) refers to deviations in the number of short tandem repeats of nucleosides in specific genomic sites that arise due to defects in the mismatch repair (MMR) mechanism, which normally detects and repairs DNA replication mistakes and maintains stability of microsatellite length. MMR deficiency (dMMR) results in MSI-high (MSI-H) tumours which are characterised by high tumour mutational burden (TMB). MSI-H/dMMR tumours produce neoantigens that increase their responsiveness to immune checkpoint inhibitors.[100,101] This is supported by evidence of high T lymphocyte infiltrates and upregulation of immune checkpoints, including programmed cell death 1 (PD-1) receptor and programmed cell death 1 ligand (PD-L1), in the microenvironment.[102] Current clinical guidelines recommend universal testing for this phenotype in all patients with mCRC to guide treatment decisions; this extends beyond those being screened due to clinical features for Lynch syndrome.[45] Although approximately 17% of early stage CRC display dMMR, this is a protective feature; hence only 4-5% of patients with mCRC display this

phenotype. Of these, one third also harbour a *BRAF* mutation. Compared with patients with proficient MMR tumours (pMMR), those with dMMR have a substantially worse prognosis which is partly driven by the higher proportion of *BRAF* mutation. [74,103]

There is now compelling evidence that patients with MSI–H/dMMR mCRC should be considered for an anti PD-1 Ab +/- CTLA-4. The phase 2 KEYNOTE-016 study evaluated the activity of anti-PD-1 checkpoint inhibitor pembrolizumab in treatment-refractory pMMR or dMMR mCRC and dMMR non-colorectal cancer. [101] The ORR in patients with dMMR mCRC was 52%, including complete responses in 12% of patients, compared to ORR of 0% in pMMR mCRC.[104] Patients with MSI-H/dMMR mCRC also had longer PFS (not reached vs 2.2 months, HR 0.10, $P<0.001$) and OS (not reached vs 5.0 months, HR 0.22, $P=0.05$) than patients with pMMR mCRC. Patients with dMMR non-colorectal cancer had similarly high response rates (ORR 71%). On the basis of these results, pembrolizumab was granted accelerated approval by the United States Food and Drug Administration (FDA) for treatment of advanced solid tumours with MSI-H or dMMR, representing the first time the agency has given tumour site-agnostic approval based on a common biomarker rather than the tumour's location of origin.

In the phase II KEYNOTE-164 study, pembrolizumab demonstrated efficacy in MSI-H/dMMR mCRC regardless of whether treatment is given as second or later line therapy. This study enrolled 124 patients following failure of $\geq$ 2 (cohort A), or $\geq$ 1 prior line of therapy (cohort B). The ORR was 33% in both cohorts. After a median follow-up of 31.3 months, the PFS was 2.3 months and 4.1 months. Median OS was 31.4 months and not reached in cohorts A and B respectively. [105] A phase III trial is currently under way comparing pembrolizumab versus standard of care chemotherapy in the first-line setting in patients with dMMR or MSI-high mCRC (NCT02563002).[106]

In the phase II CheckMate-142 study, patients with treatment-refractory dMMR/MSI-H mCRC were treated with single agent nivolumab, an alternate PD-1 immune checkpoint inhibitor, achieving an ORR of 31%.[107] Patients treated with combination immunotherapy with nivolumab and

ipilimumab (a cytotoxic T-lymphocyte-associated-4 [CTLA-4] immune checkpoint inhibitor) achieved ORR 55% and DCR 80%, at the expense of a high rate (32%) of grade 3-4 treatment-related adverse events (AE).[108] The PD-L1 inhibitor atezolizumab was tested with bevacizumab in 10 patients with MSI-H mCRC in a phase Ib study, resulting in an ORR of 30% and grade 3-4 AE rate of 40%.[109] The current evidence suggests the use of immune checkpoint inhibitors following failure of at least one line of chemotherapy however the optimal use and sequencing of agents with chemotherapy has not been ascertained.. The impact of concurrent *BRAF* mutations on efficacy of immune checkpoint inhibition is uncertain.

Atezolizumab is currently being tested in the early colon cancer setting in the ATOMIC/Alliance A021502 trial, where atezolizumab/FOLFOX is being compared to FOLFOX alone in patients with MSI-H/dMMR stage III colon cancer (NCT02912559).[110] The POLEM study is evaluating the addition of anti-PD-L1 agent avelumab in stage III colon cancer with randomised patients receiving either CAPOX/capecitabine followed by avelumab compared to chemotherapy alone (NCT03827044). [111]

Given that only a minority of CRC patients have dMMR as a biomarker predicting response to checkpoint inhibitors, there is substantial interest in developing methods to transform immunologically “cold” tumours into “hot” ones that will benefit from immunotherapy.[112] One strategy is to combine immunotherapy with molecularly targeted therapy, which can produce favourable immune effects in the tumour microenvironment such as increased antigen and HLA expression, increased T cell infiltrate, reduced immunosuppressive cytokines and improved T cell function.[113] In a phase 1b trial of the MEK inhibitor cobimetinib and atezolizumab in patients with pre-treated microsatellite stable (MSS) mCRC, 17% of patients had an objective response. [114] However, in the phase III IMblaze370 study, the combination of cobimetinib and atezolizumab failed to demonstrate improved overall survival in comparison to regorafenib. [115] The effect of chemotherapy and anti-angiogenic agents on the tumour immune milieu was examined in an early

phase trial of FOLFOX, bevacizumab and atezolizumab as first-line therapy in mCRC, resulting in ORR of 52%.[116] However, by contrast, the addition of atezolizumab to 1st line maintenance chemotherapy (fluoropyrimidine + bevacizumab), in the phase III MODUL trial did not result in an improvement in PFS, OS or ORR. [117]

Furthermore, CD8+ T cells and PD-L1 expression were increased in tumours following administration of FOLFOX, as well as after combined administration of all 3 agents, suggesting that cytotoxic chemotherapy and/or VEGF-targeted agents may help promote immune-related activity in mCRC, potentiating the efficacy of immune checkpoint inhibitors. In a phase II trial evaluating the combination of FOLFOX chemotherapy and pembrolizumab in patients with untreated mCRC, irrespective of MMR status, objective responses were reported in 16 of 30 patients (53%). [118] The PD-L1 checkpoint inhibitor atezolizumab has been tested with CEA CD3 TCB, a novel T-cell bispecific antibody targeting CEA on tumours cells and CD3 on T cells, in a phase 1a/1b trial of patients with advanced CEA-positive, chemorefractory CRC, producing responses in 1 of 5 patients receiving this combination. [119]

5.3 *HER2* amplified tumours

Human epidermal growth factor (HER2; also known as ERBB2) is a member of the epidermal growth factor (EGFR) family of transmembrane tyrosine kinase receptors, which are involved in various cellular functions such as proliferation, apoptosis, adhesion, migration and differentiation. Case series have demonstrated HER2 amplification is associated with anti-EGFR therapy resistance. [120-122] HER2-targeted agents, such as trastuzumab, are well-established as standard-of-care in *HER2*-amplified breast cancer. Based on this and evidence of activity with combination HER2-targeted agents in mouse models of *HER2*-amplified mCRC, the HERACLES trial was proof-of-concept phase 2 trial assessing the efficacy of trastuzumab plus lapatinib in HER2-positive, *KRAS* exon 2 WT mCRC patients resistant to standard therapies including cetuximab.[123] After screening 914 patients with *KRAS* exon 2 WT mCRC, 48 HER2-positive patients (defined as having either 3+ HER2 score by

immunohistochemistry (IHC), or 2+ HER2 score by IHC and fluorescence in-situ hybridisation (FISH) positive in >50% of cells) were identified and 27 were treated with dual-targeted anti-HER2 therapy. Eight patients (30%) achieved an objective response, with 7 of 8 responses seen in patients with tumours with HER2 IHC 3+ (as opposed to HER2 IHC 2+ and FISH positivity). The combination was well-tolerated, with toxicities limited to mainly grade 2 diarrhoea, fatigue and rash. The results of this study provide evidence to support the investigation of HER2-targeted therapy in earlier lines of treatment in patients with HER2-positive mCRC. In addition these data also raise questions over the implication of HER2 positivity and anti-EGFR therapy, given the potential for resistance to these agents. Of note, HER2 overexpression occurs more commonly in left-sided colon and rectal tumours than right-sided colon tumours, and thus may have significant clinical relevance for treatment choice.[124]

Oncogenic somatic mutations in *HER2* in CRC have been reported by the TCGA and occur in similar frequency to *HER2* amplification. Activating mutations involving the kinase domain (V842I, V777 and L755S) and extracellular domain (S310F) are identical to those detected in patients with breast cancer. [125] In contrast to other tumour types, in the SUMMIT basket trial of solid tumours with *HER2*/*HER3* mutations, there were no responses amongst patients with *HER2* mutated mCRC (0/12) with the pan-ERBB small molecule inhibitor neratinib. [126]

5.4 *NTRK* Fusion

Oncogenic gene fusions involving the neurotrophic tropomyosin receptor kinase (*NTRK*) occur in numerous adult malignancies [127-130], and have emerged as a promising therapeutic target. NTRK1, NTRK2 and NTRK3 (also known as TRKs) are a family of receptor tyrosine kinases involved in neuronal development. [131] The protein product of *NTRK* gene fusions is constitutively active and results in cell growth, proliferation and survival pathway activation [128-130]. The estimated frequency of *NTRK* gene fusions varies between tumour types ranging from a prevalence of >90% in

some rare cancers (secretory carcinoma of the breast and salivary glands and infantile fibrosarcoma) to <1% in more common cancer types. [128-130]

Approximately 2.5-4%, CRCs harbour these arrangements and are associated with right sided, *RAS* WT and MSI-H cancers [132,133]. Prognostically, these tumours have a poorer overall survival [133]. The gold standard for detecting these rare arrangements is reverse transcriptase polymerase chain reaction or (FISH), however immunochemistry has shown some promise as a cost-effective screening tool. [134]

Early phase studies have confirmed *NTRK* rearrangements as a targetable molecular alteration. In a pooled analysis of 3 phase I studies (NCTO2122913, SCOUT, and NAVIGATE) enrolling of the selective NTRK inhibitor larotrectinib in a tumour agnostic cohort of *NTRK* fusion positive cancers including mCRC, the ORR was 75%. Updated data together with results of an additional 67 treated patients were subsequently reported maintaining the very high response rate (now 81%, including 17% CR) with median progression-free survival of 28.3 months in the primary dataset. [135] The most common tumour types were soft tissue sarcoma (n=36), infantile fibrosarcoma (n=29), salivary gland cancer (n=21) and lung (n=12). An objective response was observed in a colorectal cancer patient. Adverse events were primarily grade 1-2, with 13% of patients having had a grade 3-4 event and only one discontinuation due to an AE related to larotrectinib.[136] The treatment was well tolerated with no grade 3/4 treatment related adverse event occurring in >5% of patients. [137]

Similar results have been reported with entrectinib in a pooled analysis of 119 patients across STARTRK-1 and ALKA-372-001 phase 1 studies [138] with an ORR of 57%, median PFS of 11.2 months, and median OS of 20.9 months. Notably high response rates were also observed in patients with intracranial metastases. [139]

Two other TRK inhibitors (repotrectinib, and LOXO-195) are also in early clinical development, particularly for patients who have progressed or are intolerant of prior TRK inhibitor therapy and

responses have reported in colorectal cancer patients with who have developed resistance whilst on treatment with larotrectinib. [140,141]

5.5 *KRAS* Mutation: G12C

Targeting KRAS has until now proved extremely difficult due to (i) RAS being a small GTPase, whose affinity for GTP exponentially exceeds that observed between kinases and ATP, and (ii) it is a small, smooth protein, with no good 'pockets' for small molecules to bind [142]. However, the KRAS G12C mutant has a cysteine residue that has been exploited to design covalent inhibitors that have promising preclinical activity. As expected, there is no preclinical activity in non- G12C mutant *KRAS* alleles such as G12D and G12V. [143]

KRAS G12C mutations are most prevalent in lung (7%) colorectal cancer (4%) and pancreatic cancer (2%) [144]. AMG510 is a novel engineered targeted agent, exploiting the substituted cysteine residue in the KRAS G12C protein. [145] Encouraging pre-clinical activity led to a phase I trial focusing on patients with previously treated *KRAS* G12C-mutant solid tumours. Initial data on the first 23 patients were presented at World Lung Conference in mid-2019 reporting a 52% ORR (and 100% disease control rate in 23 patients) [146], and at the 2019 ESMO congress, data on 12 patients with *KRAS* G12C metastatic colorectal cancer treated at the highest dose level (960mg) were presented detailing 1 PR and 10 SD for an overall disease control rate of 92%. [147] The drug was well tolerated, with no dose limiting toxicities identified. Clinical trials have just begun with a second inhibitor, MRTX849 (Mirati Therapeutics).

6. Oligometastatic Disease: Liver or Lung Dominant

6.1 Resectable and Potentially Resectable Disease

Complete resection of oligometastatic disease predominantly from the liver or lung, with or without perioperative chemotherapy, is currently the only potentially curative treatment for mCRC, with a 5-year survival rate of 40-60%.[148-150] The use of anti-EGFR therapies in the setting of resectable

liver metastases has been associated with poorer outcomes as demonstrated in the New EPOC trial and remains controversial. [151]

Conversion therapy is systemic treatment that is given to patients with potentially resectable disease with a view to 'convert' unresectable metastases to resectability.[47] Where conversion chemotherapy is used, resection rate after neoadjuvant chemotherapy has been found to correlate with tumour response rate according to RECIST criteria. Hence conversion therapy should comprise aggressive combination chemotherapy and an appropriate biological agent to aim for maximal tumour downsizing.[152-154] The optimal regimen of pre- and post-operative chemotherapy and the timing of surgery is unclear. There are concerns with the use of bevacizumab given the risks of peri-operative complications.

6.2 Unresectable Liver Dominant mCRC

Non-surgical liver-directed therapies have been investigated in trials enrolling mCRC patients with unresectable liver metastases. Radiofrequency ablation (RFA) may have a role based on the randomised phase 2 CLOCC study which compared RFA plus chemotherapy versus chemotherapy alone 30-month OS was improved with 61.7% in the combination arm compared to 57.6% in the chemotherapy only arm and median OS of 45.6 vs 40.5 months (HR 0.57, 95% CI 0.38 to 0.88, p=0.01). [155] Patients in the RFA arm did have better baseline prognostic characteristics than the control arm, with a lower proportion of patients with ≤3 metastases and higher proportion of patients with metachronous tumours and this should be taken into account.

Selective internal radiation therapy (SIRT) is the delivery of targeted radiation to liver tumours via injection of yttrium-90-labelled resin microspheres (SIR-spheres) through the hepatic artery. SIRFLOX was a phase 3 study that assessed the safety and efficacy of the addition of SIR-spheres to first-line chemotherapy (FOLFOX ± bevacizumab) in patients with unresectable liver only or liver dominant mCRC failed to meet its primary endpoint of PFS by RECIST 1.0 was not met (10.7 with SIRT vs 10.2

months without SIRT, HR 0.93, 95% CI 0.77-1.12, *P*=0.43). [156] This negative study confirmed that improvements in liver-specific PFS conferred by the addition of SIRT do not translate to improvements in OS in first line therapy. There is currently therefore no evidence to support SIRT as an adjunct to routine first-line systemic therapy in mCRC, however, a pooled analysis of three SIRT trials suggested right sided tumours may derive benefit from this approach. [157] SIRT may have a role in later lines of treatment in patients with liver only metastases who have failed chemotherapy.[158]

6.3 Oligometastatic disease: Non Liver

For patients with oligometastatic disease within organs such as ovaries or adrenal glands, surgery in conjunction with systemic chemotherapy can provide a potentially curative option. In patients who are not suitable for a surgical approach due to performance status or surgically inaccessible tumours, stereotactic radiotherapy is a reasonable modality. [159-161]

For peritoneal metastasis, aggressive cytoreductive surgery followed by hyperthermic intraperitoneal chemotherapy (HIPEC) is controversial. Retrospective case series have suggested long term survival can be achieved in a small percentage of patients with peritoneum limited metastases with cytoreductive surgery and HIPEC, however it is not possible to conclude whether the survival advantage is due to treatment or underlying tumour biology. [162-164] The largest randomised study to date, the phase III PRODIGE-7 trial, demonstrated no survival benefit in patients receiving HIPEC with oxaliplatin compared to observation following cytoreductive surgery. [165] Cytoreductive surgery for peritoneal disease should not be considered a standard of care however, if considered should be performed in centres with vast experience in peritonectomy.

7. THERAPY OF ELDERLY PATIENTS

It is well understood that the elderly represents a diverse population where the risks versus the benefits of treatment need to be individually assessed. However, to date there remains limited

research to guide the clinician on the optimal approach in the setting of metastatic colorectal cancer.

Prior to the introduction of biological agents, FOCUS-2 was the largest randomised study investigating the optimal chemotherapy regimen in the elderly. In this 2x2 factorial designed trial, the addition of oxaliplatin to fluoropyrimidines, at a reduced dose, resulted in a trend towards improved progression free survival and increased the percentage of patients with good overall treatment utility. This novel endpoint combined efficacy, toxicity and patient reflection on how worthwhile treatment had been. Substitution of capecitabine for biweekly infusional 5FU was more toxic and not associated with any efficacy or quality of life differences. [166] Thereafter, 2 large randomised studies, AVEX [19] and MAX [18] confirmed the improved response rates and PFS by adding bevacizumab to capecitabine (see Table 1). The median age of enrolled patients was 76 and 68 respectively and bevacizumab was well tolerated.

Therefore, fluoropyrimidines plus bevacizumab have been commonly employed as first line treatment. However, given the survival advantage of EGFR antibodies in first line, researchers have investigated the efficacy of cetuximab/panitumumab alone or with fluoropyrimidine monotherapy in *RAS* WT patients. Two such ongoing studies include the AGITG MONARCC (ACTRN12618000233224) and the GONO PANDA (NCT02904031). Both have a minimum recruitment age of 70 and are incorporating geriatric assessments to assess their utility in predicting treatment outcome and chemotherapy toxicity. The MONARCC study assesses overall treatment utility and the PANDA study specifically evaluates the "CRASH score" [167] as a predictor of chemotherapy toxicity and to validate a brief, user-friendly screening tool (G 8) to select patients who need a comprehensive geriatric assessment. [168] It is hoped this type of research will provide data to support targeted incorporation of geriatric assessments into the clinic, providing the clinician with objective data which can frame discussions with the patient and family as well as be a basis for rational treatment decisions.

Certain molecular subsets are more common in the elderly. Specifically, in a large Australian database *BRAF* mutations were found in 18% of patients above 80 years versus 11% in those under 70 years [169]. The same registry found that microsatellite instability is more frequent with age such that in those over 80 years, approximately 15% were microsatellite unstable. [170] Given the results of the BEACON study and impact of immunotherapy on microsatellite unstable cancers, these trends have direct implications for therapy options in the elderly. *KRAS* mutations (but not *NRAS*) are also more common as age increases, such that the frequency of mutations rises from approximately 40% in those under 70 to 60% above the age of 80. Furthermore, specific mutations vary with age, with a greater prevalence of codon 12 (including G12C) mutations in younger patients and by contrast a greater prevalence of codon 61 (especially Q61), 117 and 146 mutations as age increases. [169,171]

8. Future Translational Research

6.1 Circulating Tumour DNA

The use of circulating tumour DNA (ctDNA) remains a research tool but is generating considerable interest, particularly around its prognostic role in early stage disease, after resection of metastatic disease and in providing real time information on resistance mechanisms as patients progress through treatment for metastatic disease. Studies have demonstrated a high degree of concordance in mutations detected in ctDNA and tumour tissue [172] and can represent a non-invasive alternative to tumour biopsy to establish the molecular status of mCRC such as MSI [173] and *RAS/RAF* mutation status. [174]

The quantity of ctDNA is proportional to tumour burden. Longitudinal measurements of ctDNA during chemotherapy show proportional decreases and increases according to clinical response. [175-177] Henceforth, ctDNA has utility to evaluate early responses to treatment. [178]

The emergence of *RAS* mutations is a mediator of acquired resistance to anti-EGFR therapy. [179] In a pivotal study, Diaz et al was able to detect the emergence of resistance conferring *KRAS* mutations in a cohort of patients receiving panitumumab. [174] Data from CRICKET study suggest the

persistence of these mutations following first-line cetuximab-irinotecan predicted resistance to cetuximab-irinotecan rechallenge in the third line setting. [180]

In the adjuvant setting, ctDNA is a prognostic biomarker for minimal residual disease. [181] There is ongoing investigation into the utility of stratification of risk and refining the adjuvant chemotherapy, based on the presence or absence of ctDNA (NCT04050345, ACTRN12617001566325). These studies will go a long way to informing the role of ctDNA in clinical practice as well as the optimal technological platforms for ctDNA detection.

6.2 Gene Expression Signature-defined Molecular Subgroups

Gene expression signatures are of interest as tools to refine classification of CRC and facilitate clinical prognostication and development of expression signature-based targeted therapies. The Consensus Molecular Subtypes (CMS) is most mature and was developed by the CRC Subtyping Consortium.[182] The CMS system classifies CRC into 4 subtypes based on gene expression signatures. The prognostic role of CMS subtypes has been studied in a number of retrospective analyses including the MAX, CALGB 80405 and FIRE-3 studies comparing the first-line combinations of chemotherapy with EGFR- versus VEGF-targeted agents.[183-185] However, the results from these studies are discrepant and are demonstrative of the complex interplay between tumour microenvironment, chemotherapy and targeted agents. [186] The prognostic and potentially predictive value of CMS subtypes however awaits validation in prospective trials, and there is currently insufficient evidence for using CMS subgroups to inform clinical decision making.

Additionally, the gene expression signatures are not readily able to be analysed in most cancer centres, leading a focus on developing a rapid, standard test such as immunohistochemistry to determine CMS subtype.

6.3 Microbiome

The microbiome is the collection of bacteria, viruses, fungi and protozoa that reside in the human body. Given the causative link between CRC and dietary patterns which may alter the composition of

the gut microbiome, research into the microbiome for CRC prevention, diagnostics and therapeutics is ongoing. [187] Specific species such as *F. nucleatum [188]*, *B. fragilis*, and *E.coli* [189] play may a role in carcinogenesis and cancer progression by multiple mechanisms including promotion of inflammation and secretion of biofilms. Currently, there are no approved therapies targeting the microbiome however, this field represents an opportunity for research. Understanding the complex interaction between the colon and the microbiome may unlock new biomarkers or therapeutic interventions leading to tumour control or toxicity management.

9. Expert Opinion

This consensus review examines current evidence for the use of targeted therapy in mCRC in different settings. There is well-established evidence for the use of EGFR-targeted and VEGF-targeted antibodies, which should routinely be incorporated into treatment strategies for mCRC. The use of EGFR-targeted antibodies should be restricted to patients with extended *RAS* WT profiles (*NRAS* 2-4 WT and *KRAS* 2-4 WT). For this group of patients, the choice of bevacizumab versus anti-EGFR therapy in the first-line setting now appears clearly based on side of primary. Based on retrospective analyses of randomised trials, left-sided tumours appear to derive greater benefit from EGFR-targeted antibodies compared to right-sided tumours. Therefore, primary tumour location should be taken into consideration when selecting biological treatments, which is a recommendation that is already reflected in some colorectal cancer treatment guidelines.

Novel subgroups and classification systems are gaining interest as tools to aid better characterisation, prognostication and development of targeted therapies in mCRC. While *BRAF* mutations in mCRC are associated with poor prognosis and less likelihood of progressing to subsequent treatment lines, these mutations represent a potential target for biological treatment. Novel treatment combinations of BRAF and MEK inhibitors particularly in combination with anti-

EGFR agents have demonstrated activity in a randomised phase III trial of *BRAF* V600E mutant mCRC. MMR deficiency is recognised as being predictive of benefit from immune checkpoint inhibitors, which have now been approved for use in later lines in this molecular subgroup, not only in mCRC but across all solid tumours but the exact timing of anti PD-1/IO agents +/- combination IO, await the outcome of completed phase III trials. *HER2* amplifications and *NTRK* fusions also represent an important target for developing additional treatments in mCRC and highlight the importance of identification of rare molecular subgroups. In terms of novel treatment modalities, inhibitors of KRAS G12C are promising and represent a major advance in an oncogene which was previously thought to be untargetable.

Finally, translational research such as ctDNA and CMS subgroups represents an opportunity for further research and personalisation of patient care. Further research into the tumour microbiome may unlock new insights and treatments in mCRC.

ARTICLE HIGHLIGHTS

- The treatment strategy for advanced colorectal cancer should include early molecular assessment and multidisciplinary review to determine the optimal systemic options and also the potential for resection of metastasis.
- Site of primary is now accepted by a number of guidelines (NCCN, ESMO and Australian NHMRC) as a guide to treatment choice in *RAS* WT mCRC, with left-sided tumours best treated with an anti-EGFR/chemotherapy combination, and right-sided tumours with bevacizumab/chemotherapy combination
- Mismatch repair (MMR) status should be routinely tested in mCRC patients as MMR deficiency predicts benefit from immune checkpoint inhibitors in the treatment-refractory setting with the question remaining re timing and single agent versus combinations.
- Patients with *BRAF* mutations should be considered for combination therapy with anti-EGFR/BRAF +/-MEK; the optimal sequencing with other therapies remains unclear

- *HER2* amplifications may be amenable to treatment with molecularly targeted agents, pending further clinical trial evidence.
- New targets such as *KRAS* G12C and *NTRK* fusions may lead to further options and improved outcomes.
- Circulating tumour DNA (ctDNA) allows real time tumour mutational and burden monitoring and may better inform clinical decision making.
- Currently there is evidence that mCRC could be divided into at least 6 distinct clinical/molecular subgroups which have distinct treatment pathways; 1. Left sided *RAS* WT, 2. Right sided *RAS* WT, 3. *RAS* MT, 4. *BRAF* MT, 5. HER2 over expressed, and 6. dMMR (noting some cross over with *BRAF* MT).

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Table 1: First-line Phase III trials with anti-angiogenic agents

BIOLOGIC	Treatment	n	ORR (%)	Median PFS months (HR)	Median OS months (HR)	Comment
Bevacizumab [16]	IFL V IFL/ bev	813	35 V 45 +	6.2 V 10.6 (0.54) +	15.6 V 20.3 (0.66) +	No randomised data available for FOLFIRI
Bevacizumab (NO16966) [17]	Oxaliplatin/ fluoropyrimidine V Oxaliplatin/fluoropyrimidine / bev	1401	38 V 38 -	8.0 V 9.4 (0.83) +	19.9 V 21.3 (0.89) -	Suboptimal use of drugs
Bevacizumab (MAX) [18]	Capecitabine V capecitabine (mitomycin)/ bev	471	30 V 36 -	5.7 V 8.5 (0.63) +	18.9 V 16.4 -	
Bevacizumab (AVEX) [19]	Capecitabine V capecitabine/bev	280	10 V 19 +	5.1 V 9.1 (0.53) +	16.8 V 20.7 (0.78) -	Cape/bev more toxic but well tolerated in elderly (age >70 years)
Bevacizumab +/- Panitumumab (PACCE) [49]	Doublet chemotherapy /bev V Doublet chemotherapy/bev/pan	823	48 V 46 -	11.4 V 10.0 (1.27) -	24.5 V 19.4 (1.43) -	Combined Abs inferior Trend to inf ORR, PFS , OS in *KRAS* WT
Bevacizumab +/- cetuximab (CAIRO2) [25]	Oxaliplatin/capecitabine/bev V Oxaliplatin/capecitabine/bev/cet	736	50 V 52.7 -	10.7 V 9.4 (1.22) +	20.3 V 19.4 -	Combined Abs inferior

Bevacizumab (TRIBE) [12]	FOLFIRI/bev V FOLFOXIRI/bev	508	54 V 65	9.7 V 12.3 (0.77) +	25.8 V 29.8 (0.80) +	OS in *RAS*/*BRAF* WT and *RAS*/ *BRAF* mutated subgroups presented in update
Bevacizumab (ITACa) [190]	Doublet chemotherapy V Double chemotherapy/bev	376	50.0 V 50.6 -	8.4 V 9.6 (0.86) -	21.3 V 20.8 (1.13) -	

NB + is statistically significant, - is not, ORR=objective response rate, HR=hazard ratio, PFS=progression free survival , OS =overall survival, bev=bevacizumab, cet=cetuximab, pan=panitumumab, WT=wild type

Table 2. Maintenance phase III trials with anti-angiogenic agents

Biologic	**First-line Induction Treatment**	**Treatment**	**n**	**Median PFS months (HR)**	**Median OS months (HR)**	**Comment**
Bevacizumab (MACRO) [22]	18 weeks CAPOX/ bev	CAPOX/bev V bev	480	10.4 V 9.7 (1.10) -	23.2 V 20.0 (1.05) -	Non-inferiority endpoint for PFS not met Randomisation occurred before induction treatment
Bevacizumab (CAIRO3) [23]	18 weeks CAPOX/bev	Capecitabine/bev V Observation	558	8.5 V 4.1 (0.40) +	25.9 V 22.4 (0.83) -	Positive for primary endpoint of time to second progression (HR 0.67, p<0.0001)

						Randomisation occurred after induction treatment
Bevacizumab (PRODIGE 9) [24]	24 weeks FOLFIRI/bev	Bevacizumab V Observation	494	9.2 V 8.9 (0.92) -	21.65 V 21.98 (1.05) -	Randomisation occurred before induction treatment
Bevacizumab (AIO 0207) [191]	24 weeks Fluoropyrimidine/oxaliplatin/ bev	Fluoropyrimidine/bev V Bev V Observation	837	6.3 V 4.6 V 3.5 +	20.2 V 21.9 V 23.1 -	Non-inferiority demonstrated for primary endpoint (failure of strategy) for bev compared to fluoropyrimidine/bev (HR 1.08, 95% CI 0.85-1.37) Randomisation occurred after induction treatment
Bevacizumab/ erlotinib (GERCOR DREAM; OPTIMOX 3) [26]	24 weeks Fluoropyrimidine/oxaliplatin/ bev or FOLFIRI/bev	Bev/erlotinib V Bev	700	5.4 V 4.9 (0.81) -	24.9 V 22.1 (0.79) +	Randomisation occurred after induction treatment

NB + is statistically significant, - is not, ORR=objective response rate, PFS=progression free survival,

OS =overall survival, HR=hazard ratio, bev=bevacizumab, cet=cetuximab, pan=panitumumab

Table 3: Phase III Trials adding EGFR antibodies to chemotherapy

FIRST-LINE	TREATMENT		*KRAS* exon 2 wild type analysis				Extended *RAS* wild type analysis			
			n	ORR %	PFS m; (HR)	OS m; (HR)	n	ORR %	PFS m; (HR)	OS m; (HR)
Cetuximab (CRYSTAL)	FOLFIRI/cetuximab V FOLFIRI		666	57 V 40 +	9.9 V 8.4 (0.696) +	23.5 V 20.0 (0.796) +	367	66 V 39 +	11.4 V 8.4 (0.56) +	28.4 V 20.2 (0.69) +
Panitumumab (PRIME)	FOLFOX/panitumumab V FOLFOX		656	57 v 48 +	9.6 V 8.0 (0.80) +	23.9 V 19.7 (0.83) -	512	NR	10.1 V 7.9 (0.72) +	26 V 20.2 (0.78) +
Cetuximab (NORDIC)	Nordic FLOX/Cetuximab V Nordic FLOX		303	46 v 47 -	7.9 V 8.7 -	20.1 V 22.0 -	N/A	N/A	N/A	N/A
Cetuximab (COIN)	Oxaliplatin/FU/ cetuximab v oxaliplatin/FU		729	64 v 57 +	8.6 V 8.6 -	17.0 V 17.9 -	N/A	N/A	N/A	N/A

SECOND-LINE	**TREAMENT**	**Prior Bev**	***KRAS* exon 2* wild type analysis**				**Extended *RAS* wild type analysis**			
			n	ORR%	PFS HR	OS HR	n	ORR%	PFS HR	OS HR
Panitumumab (PICCOLO)*	Irinotecan vs Irinotecan + panitumumab	2% 2%	460 (KRAS exons 2 and 3)	12 V 33 +	0.78 +	1.01 -	323	12 V 44 +	0.68 +	0.92 -
Panitumumab 20050181	FOLFIRI V FOLFIRI/ panitumumab	20% 18%	597	10% V 35% +	0.73 +	0.85 -	421	10% V 41% +	0.70 +	0.81 -
Cetuximab (Study CA225006)	Irinotecan V Irinotecan/ cetuximab	N/A N/A	192	N/A	0.773 -	1.29 -	N/A	N/A	N/A	N/A

* patients in the PICCOLO study were wild type for *KRAS* exons 2 and 3

NB + is statistically significant, - is not, ORR=objective response rate, PFS=progression free survival, OS =overall survival, HR=hazard ratio, NR= not reached,

N/A = not applicable

Table 4: Head –to- head first-line trials comparing anti-EGFR therapies with bevacizumab

Trial	Treatment	***KRAS* exon 2 wild type analysis**				**Extended *RAS* wild type analysis**			
		n	**ORR %**	**PFS Months (HR)**	**OS Months (HR)**	**n**	**ORR %**	**PFS Months (HR)**	**OS Months (HR)**
PEAK Phase II	FOLFOX + bev V FOLFOX + pan	285	54 V 58	10.1 V 10.9 (0.87) -	24.3 V 34.2 (0.62) +	170	60 V 64	9.5 V 13.0 (0.65) +	28.9 V 41.3 (0.63) -
FIRE3 Phase III	FOLFIRI/bev V FOLFIRI/cet	592	58 V 62 -	10.0 V 10.3 (1.06) -	25.0 V 28.7 (0.77) +	342	60 V 66 -	10.2 V 10.4 (0.93) -	25.6 V 33.1 (0.70) +
CALGB80405 Phase III FOLFOX or	Doublet/bev V	1137	55.2 V 59.6	10.6 V	30.0 V	526	56 V	11.0 V	31.2 V

FOLFIRI doublet backbone (73% FOLFOX)	Doublet/cet		-	10.5 (0.95) -	29.0 (0.88) -		68.8 +	11.2 (1.03) -	32.0 (0.88) -

NB + is statistically significant, - is not, ORR=objective response rate, PFS=progression free survival, OS =overall survival, HR=hazard ratio, NR= not reached,

N/A = not applicable, bev=bevacizumab, pan=panitumumab, cet=cetuximab

Table 5: Second-line trials with anti-angiogenic agents

	Treatment	N	Prior Bevacizumab	PFS Months (HR)	OS months (HR)
Aflibercept (VELOUR)	FOLFIRI + aflibercept V FOLFIRI + placebo	612 614	30.4% 30.5%	6.9 V 4.7 (0.758) +	13.5 V 12 (0.82) +
Bevacizumab (ECOG 3200)	FOLFOX + bevacizumab V FOLFOX	293 292	0% 0%	7.3 V 4.7 (0.61) +	12.9 V 10.8 (0.75) +
Bevacizumab (ML18147)	Oxali-/Iri-CT + bevacizumab V Oxali-/Iri- CT alone	409 411	100% 100%	5.7 V 4.1 (0.68) +	11.2 V 9.8 (0.81) +
Bevacizumab (BEBYP)	mFOLFOX-6/FOLFIRI + bevacizumab V mFOLFOX/FOLFIRI	92 92	100% 100%	6.8 V 5.0 (0.70) +	14.1 V 15.5 (0.77) +
Ramucirumab (RAISE)	FOLFIRI + ramucirumab V FOLFIRI	536 536	100% 100%	5.7 V 4.5 (0.793) +	13.3 V 11.7 (0.844) +

NB + is statistically significant, - is not, HR=hazard ratio, PFS=progression free survival , OS =overall survival, Oxali=oxaliplatin, Iri=irinotecan, CT=Chemotherapy