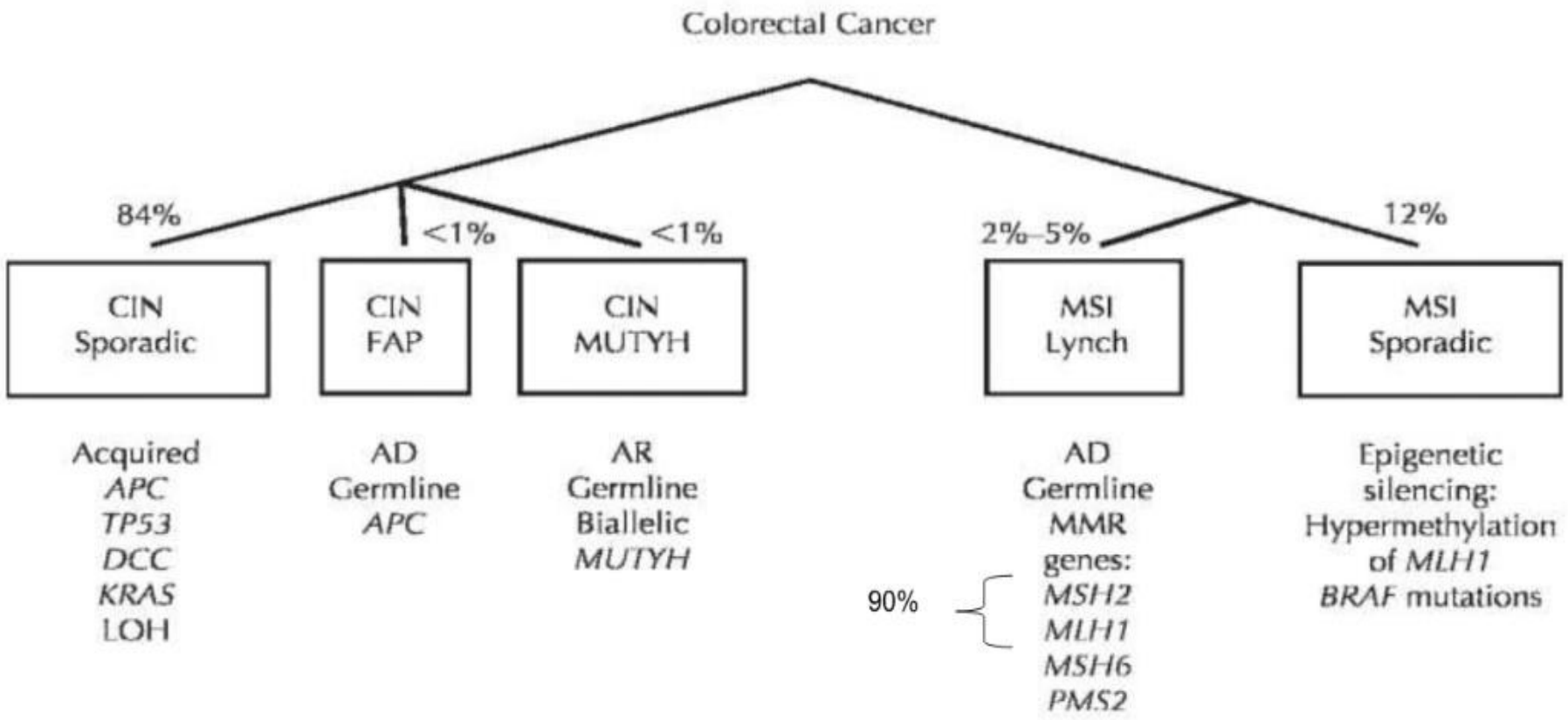
Mohammadreza Khosravi

Hematologist and medical oncologist

Types of CRC

- **Sporadic =** 70% of cases
  - there is no family history, common above age of 50
- **Inherited** = *1 – 6%* of cases
  - Hereditary NonPolyposis CRC (HNPCC, Lynch syndrome)
  - Multiple polyps CRC
    - familial adenomatous polyposis (FAP)
    - hamartomatous polyposis syndromes (e.g. Peutz-Jeghers, juvenile polyposis)
    - MUTYH-associated polyposis (MAP)
- **Familial CRC = 25%** of cases
  - have a family history of CRC, but the pattern is not consistent and the risk is not as high as with the inherited syndromes.
    - single affected first-degree relative = 1.7 ↑^{ed} risk
    - two affected first-degree relatives or if Dx before age 55 = Further ↑ risk


Colorectal Cancer
84%
<1%
<1%
2%–5%
12%
CIN Sporadic
CIN FAP
CIN MUTYH
MSI Lynch
MSI Sporadic
Acquired
APC
TP53
DCC
KRAS
LOH
AD
Germline
APC
AR
Germline
Biallelic
MUTYH
AD
Germline
MMR
genes:
MSH2
MLH1
MSH6
PMS2
90%
Epigenetic
silencing:
Hypermethylation
of MLH1
BRAF mutations

Molecular biomarkers in oncology

There is no single agreed definition of personalized medicine

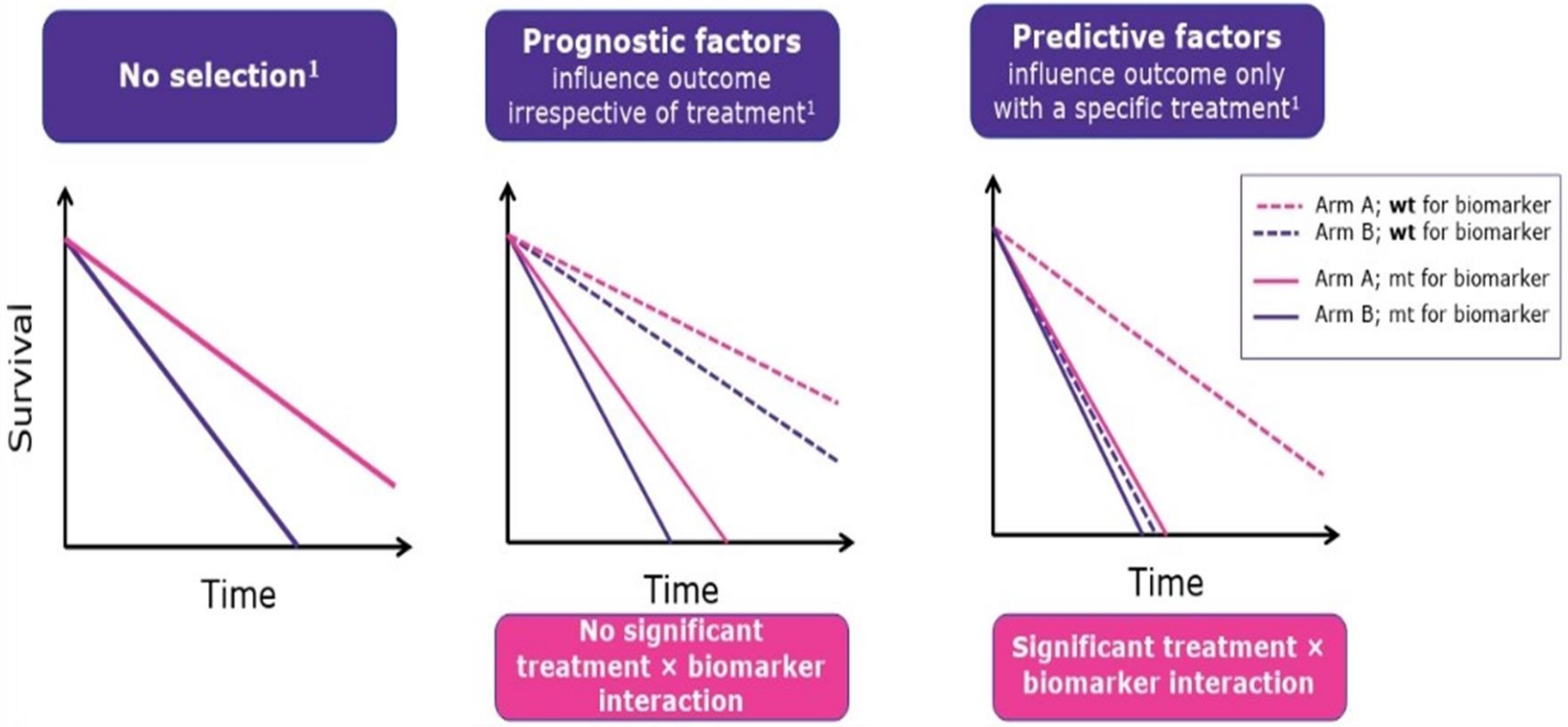
"A medical model using molecular profiling for tailoring the right therapeutic strategy for the right person at the right time, and/or to determine the predisposition to disease and/or to deliver timely and stratified prevention"
(European Commission)[1]

"The application of genomic and molecular data to tailor medical care to individuals"
(National Cancer Institute)[2]

Information about variations, mutations or changes in expression of key proteins, or epigenetic changes, may be prognostic or predictive, or both

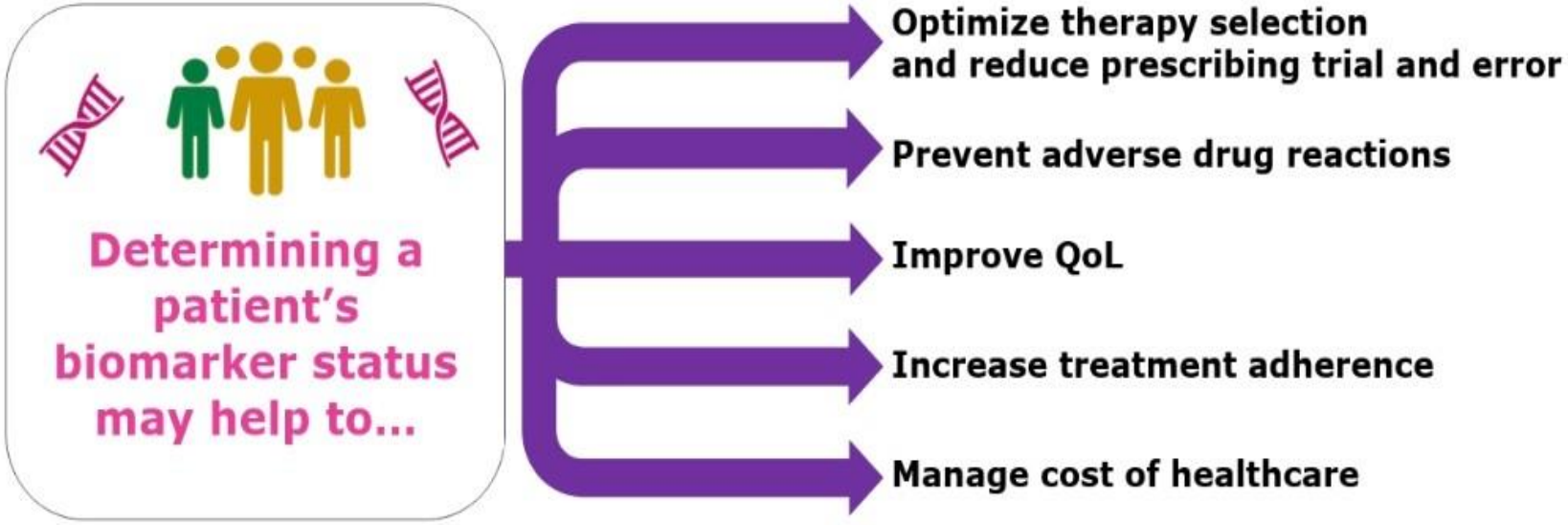
Molecular biomarkers in oncology

Biomarkers may be prognostic or predictive, or both



Molecular biomarkers in oncology

Personalized medicine has a number of benefits for patients



Noninvasive Biomarkers of Colorectal Cancer: Role in Diagnosis and Personalised Treatment Perspectives

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Colorectal cancer (CRC) is the third leading cause of cancer-related deaths worldwide. It has been estimated that more than one-third of patients are diagnosed when CRC has already spread to the lymph nodes. One out of five patients is diagnosed with metastatic CRC. The stage of diagnosis influences treatment outcome and survival. Notwithstanding the recent advances in multidisciplinary management and treatment of CRC, patients are still reluctant to undergo screening tests because of the associated invasiveness and discomfort (e.g., colonoscopy with biopsies). Moreover, the serological markers currently used for diagnosis are not reliable and, even if they were useful to detect disease recurrence after treatment, they are not always detected in patients with CRC (e.g., CEA). Recently, translational research in CRC has produced a wide spectrum of potential biomarkers that could be useful for diagnosis, treatment, and follow-up of these patients. The aim of this review is to provide an overview of the newer noninvasive or minimally invasive biomarkers of CRC. Here, we discuss imaging and biomolecular diagnostics ranging from their potential usefulness to obtain early and less-invasive diagnosis to their potential implementation in the development of a bespoke treatment of CRC.

Table 1

Potential applications of LB in CRC
Early diagnosis
Assessment of molecular heterogeneity of overall disease
Identification of genetic alterations for targeted therapy
Evaluation of tumor response after preoperative treatments
Monitoring of minimal residual disease
Assessment of evolution of resistance in real time

Table 2: Imaging Biomarkers.

Modality	Parameters	Application
CT	Anatomical and functional imaging (DCET-CT)	Staging and treatment response
MRI	Anatomical and functional imaging (DWI, DCE-MRI, TA)	Diagnosis, local staging, prognostic evaluation, treatment response
PET-CI	Metabolic and anatomical imaging	Diagnosis, staging and treatment response
PET-MRI	Metabolic and anatomical imaging	Diagnosis, staging, prognostic evaluation, treatment response

cancers

MDPI

Review

Prognostic and Predictive Molecular Biomarkers for Colorectal Cancer: Updates and Challenges

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check for updates

Abstract: Colorectal cancer (CRC) is a leading cause of death among cancer patients. This heterogeneous disease is characterized by alterations in multiple molecular pathways throughout its development. Mutations in *RAS*, along with the mismatch repair gene deficiency, are currently routinely tested in clinics. Such biomarkers provide information for patient risk stratification and for the choice of the best treatment options. Nevertheless, reliable and powerful prognostic markers that can identify "high-risk" CRC patients, who might benefit from adjuvant chemotherapy, in early stages, are currently missing. To bridge this gap, genomic information has increasingly gained

Table 1. Current clinical used molecular biomarkers in colorectal cancer (CRC).

Status	Prevalence	Prognostic Value	Predictive Value		Ref.
			Non-Metastatic	Metastatic	
Microsatellite instability (MSI) high	overall: 15% stage II/III: 15% stage IV: 4.5%	Increased overall survival in stage II	× 5-FU monotherapy (stage II) √ FOLFOX (stage III)	√ Immuno-therapy	[11–24]
KRAS/NRAS mutation	45%	Bad prognostic (debatable in stage II)	No impact on treatment	× Anti-EGFR therapy	[25–38]
BRAF mutation	7–10%	Bad prognostic especially in the metastatic setting	Currently no impact on treatment	× Anti-EGFR therapy (conflicting data) √ Combi therapy: Anti-BRAF/Anti-EGFR	[12–30,39–43]

Benefit: √; No benefit: ×.

Table 2. List of promising future biomarkers in CRC.

Biomarkers	Value	Therapy Involved	Target Group	Ref.
Currently Translated into Daily Routine				
ctDNA	Predictive/prognostic	√ Targeted therapies √ Chemotherapy duration	All CRC	[72–79]
Tumor sidedness	Prognostic/predictive	× Anti-EGFR therapy (RCC) √ Intensive Chemotherapy (RCC) √ Immunotherapy (RCC) (most likely)	All CRC/mCRC	[80–83]
ALK, ROS1, NTRK1-3 fusions	Predictive	√ ALK, ROS and TRK inhibitors	All CRC ALK, ROS and NTRK fusions	[84–88]
HER2 amplification	Predictive	√ Anti-HER2 strategies × Anti-EGFR strategies	All CRC with HER2 amplification	[89,90]
Need for Further Investigation				
CMS subtyping	Prognostic	Predictive value currently studied	All CRC	[12,91–94]
Immunoscore	Prognostic	Predictive value not yet determined	All CRC	[95–100]
Markers based on the stromal compartment	Prognostic/predictive	× Adjuvant chemotherapy in rectal cancer	All stages with a focus on stage II for the prognostic value	[101–105]
Tumor mutation burden	Predictive	√ Immunotherapy	mCRC	[77–79]
CIMP	Prognostic	Predictive value not yet determined	All CRC	[106–108]
PI3K	Predictive	× Anti-EGFR therapy × First-line chemotherapy	mCRC All CRC	[40,109–111]
miR-31-3p (low expression)	Predictive	√ Anti-EGFR therapy	KRAS wt mCRC	[112–116]
Promising Biomarker Specifically Focusing on Stage II CRC				
CDX2 (low expression)	Prognostic	Predictive value not yet confirmed	Stage II CRC	[117]
MYO5B (low expression)	Prognostic	Predictive value currently studied	Stage II CRC	[118]

Benefit: √; No benefit: ×.

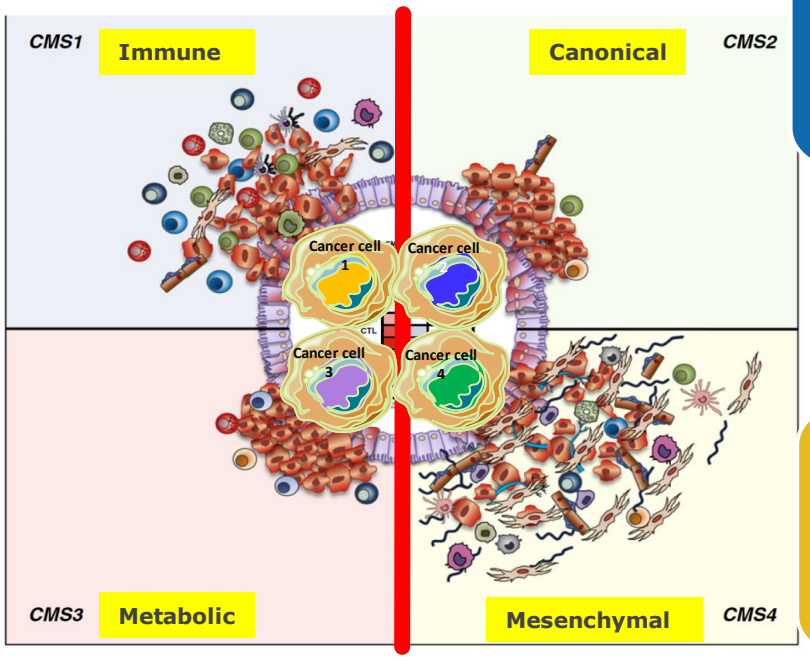
CMS classification is based on gene expression and reflects the functional implications of molecular status[27]

CMS

CMS1
High MSI, hypermutation, and immune hot micro-environment

CMS2
Epithelial phenotype, chromosomally unstable with WNT and MYC signaling activation

Distribution of TME cell populations in the four CMS subtypes[28]



CMS3
Epithelial phenotype, metabolic dysregulation

CMS4
Mesenchymal phenotype with TGF-β activation, stromal invasion, and angiogenesis

Image taken from Colangelo T, et al. Biochim Biophys Acta Rev Cancer 2017;1867:1–18.

▸TGF-β, transforming growth factor-β; TME, tumor microenvironment.

27. Aderka D, et al. Lancet Oncol 2019;20:e274–e283; 28. Colangelo T, et al. Biochim Biophys Acta Rev Cancer 2017;1867:1–18.

An effective 1L treatment combination may double the attainable mOS[27]

CMS

Median Overall Survival[27]

	CALGB/SWOG 80405*† Oxaliplatin 75%		FIRE-3*‡ Irinotecan 100%	
	Cetuximab + CT§	Bevacizumab + CT§	Cetuximab + CT§	Bevacizumab + CT§
CMS1	11.7	22.5	17.9	13.1
CMS2	42.0	36.0	38.3	29.1
CMS3	26.8	15.1	16.6	18.6
CMS4	30.8	32.7	40.1	21.1

Optimal combination for this CMS type in these studies

As a result of these interactions, cetuximab is an important option for an effective combination for 86% of KRAS wt tumors (CMS2, CMS3, and CMS4)[27]

*The OS benefit of Erbitux + CT vs bevacizumab + CT demonstrated for patients with RAS wt mCRC in the FIRE-3 study could not be confirmed in the CALGB/SWOG 80405 study.[17,18] †The CALGB/SWOG 80405 study did not meet its primary endpoint of significantly improving OS in the cetuximab + CT arm vs bevacizumab + CT arm in patients with KRAS (exon 2) wt mCRC.[17] ‡FIRE-3 did not meet its primary endpoint of significantly improving ORR based on investigators' read in patients with KRAS (exon 2) wt mCRC.[19] §CT regimens were FOLFOX/FOLFIRI.[18]

17. Venook AP, et al. JAMA 2017; 317:2392–2401; 18. Heinemann V, et al. Lancet Oncol 2014;15:1065–1075; 27. Aderka D, et al. Lancet Oncol 2019;20:e274–e283.

Consensus molecular subtyping: Overview

CMS1 MSI immune	CMS2 Canonical	CMS3 Metabolic	CMS4 Mesenchymal
14%	37%	13%	23%
MSI, CIMP high, hypermutation	SCNA high	Mixed MSI status, SCNA low, CIMP low	SCNA high
BRAF mutations		*KRAS* mutations	
Immune infiltration and activation	WNT and MYC activation	Metabolic deregulation	Stromal infiltration, TGF-β activation, angiogenesis
Worse survival after relapse			Worse relapse-free and overall survival

Guinney et. al. Nature Medicine 2015

Sidedness in a structural concept.

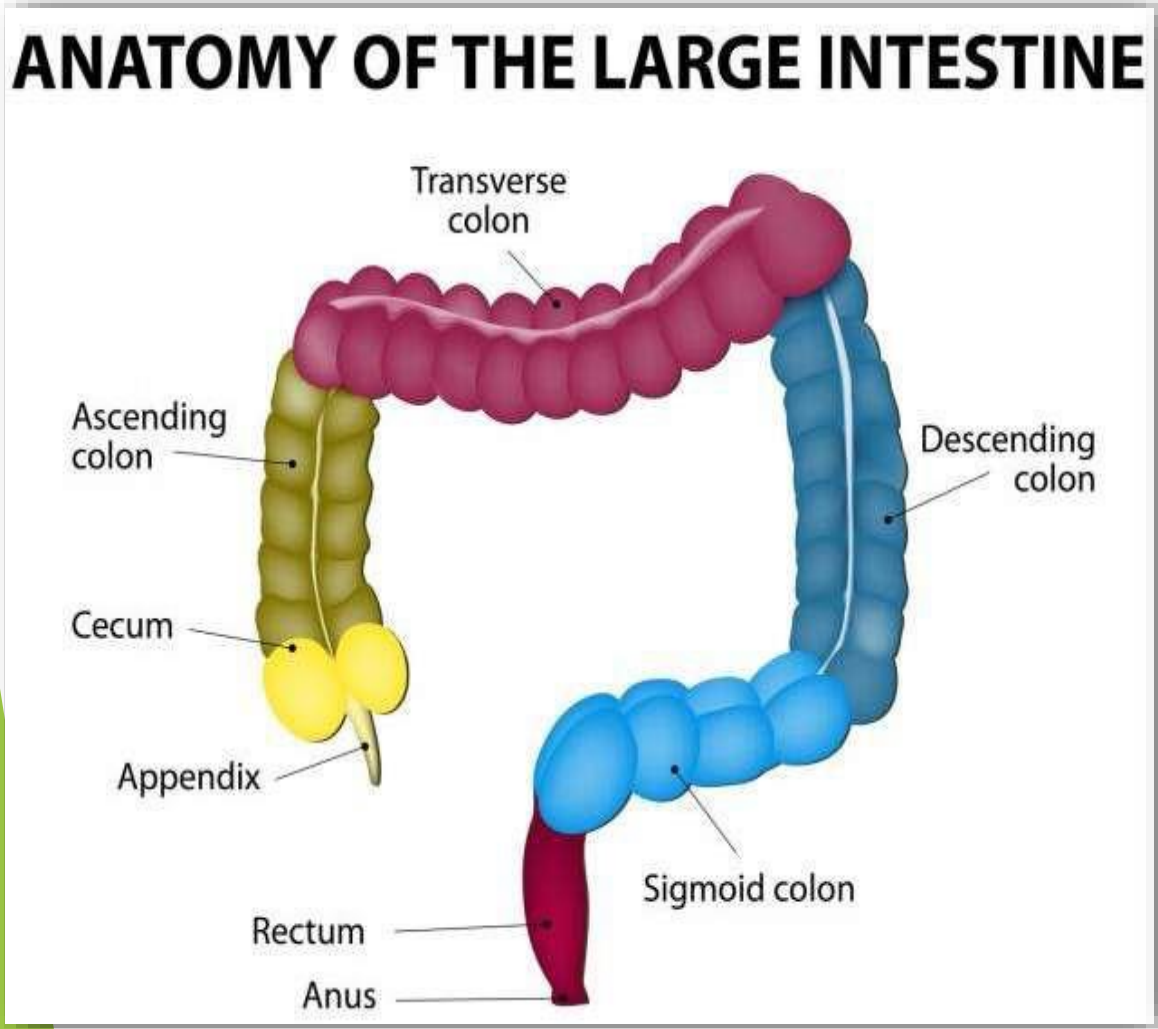


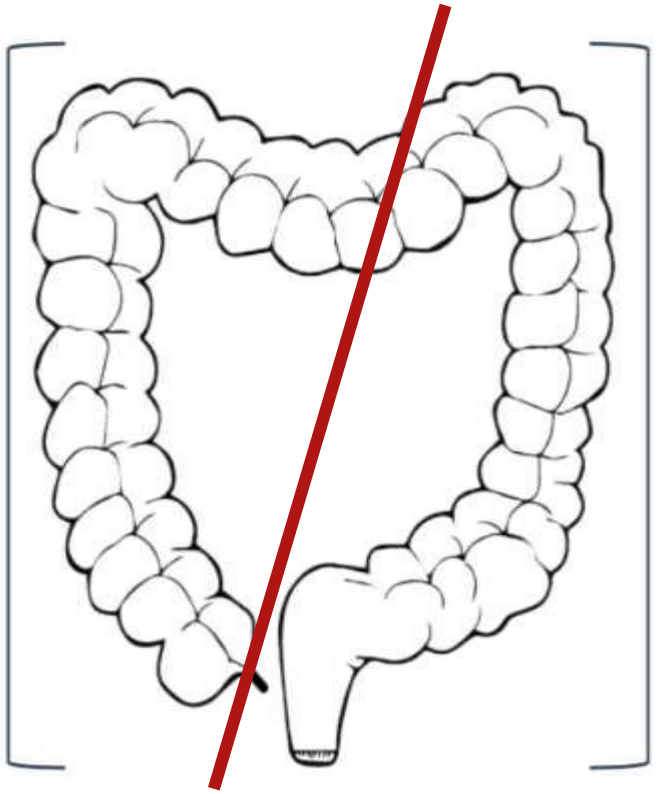
FIGURE 2. Differences in clinical factors, molecular markers, and gene expression profile of right- versus left-sided CRCs.

Right-Sided
Cecum to hepatic flexure

Clinical
- *Older age*
- *Mucinous histology*
- *Worse prognosis*

Gene Expression Profile:
- *CMS-1 (immune)*
- *CMS-3 (metabolic)*
- *Serrated pathway Signature*

Molecular Markers:
- *dMMR/MSI-H*
- CIMP-high
- *BRAF mutations*
- *EREG-, AREG-low*



Left-Sided
Rectum to splenic flexure

Clinical
- *Younger age*
- *Better prognosis*

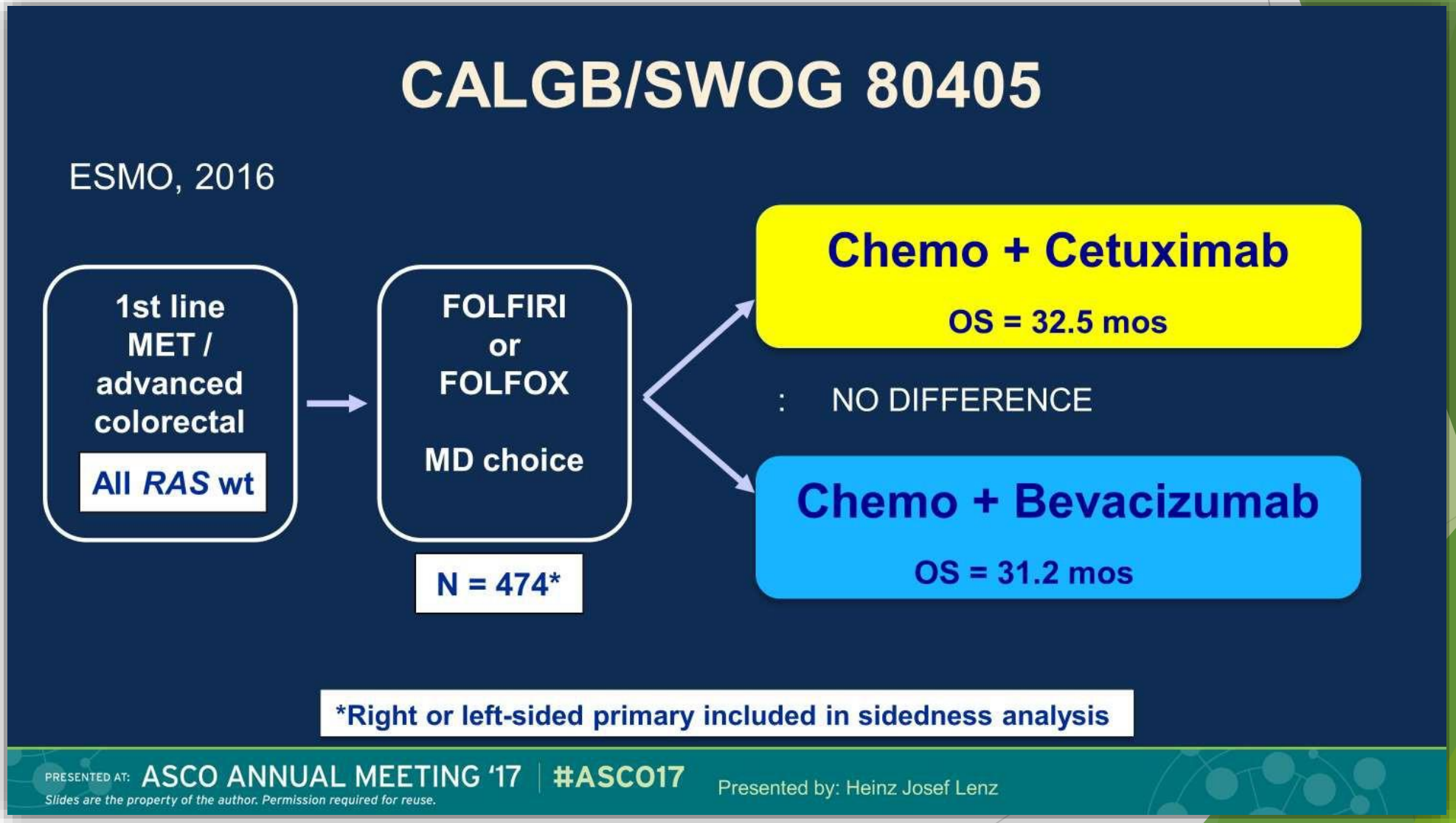
Gene Expression Profile:
- *CMS-2 (canonical)*
- *CMS-4 (mesenchymal)*

Molecular Markers:
- *Classic chromosomal instability*
- CIMP-low
- *EREG-, AREG-high*

5-FU = 5-fluorouracil; ASCO = American Society of Clinical Oncology; GI = gastrointestinal; IFL = irinotecan/fluorouracil/levoleucovorin; OS = overall survival; PFS = progression-free survival; WT = wild type.

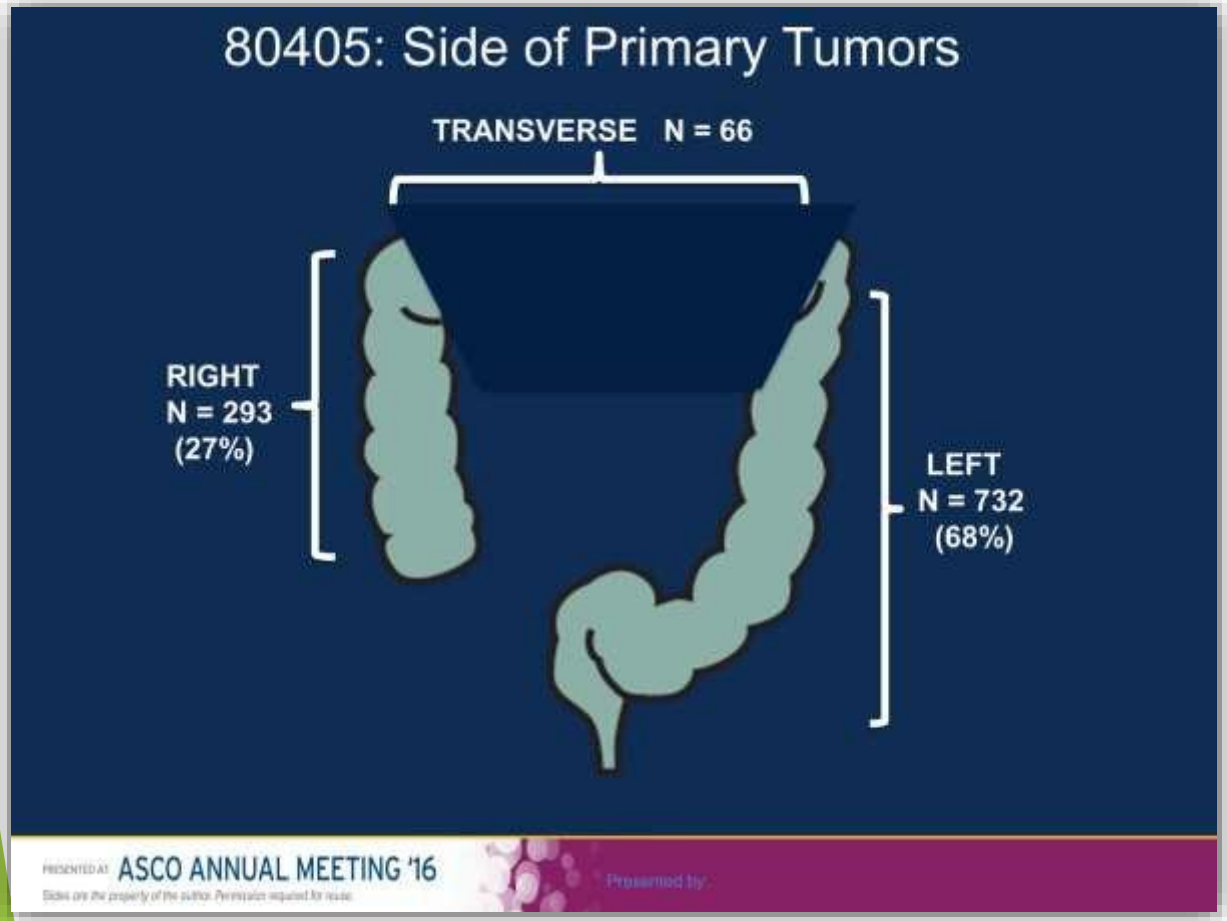
King et. al. AJHO 2016

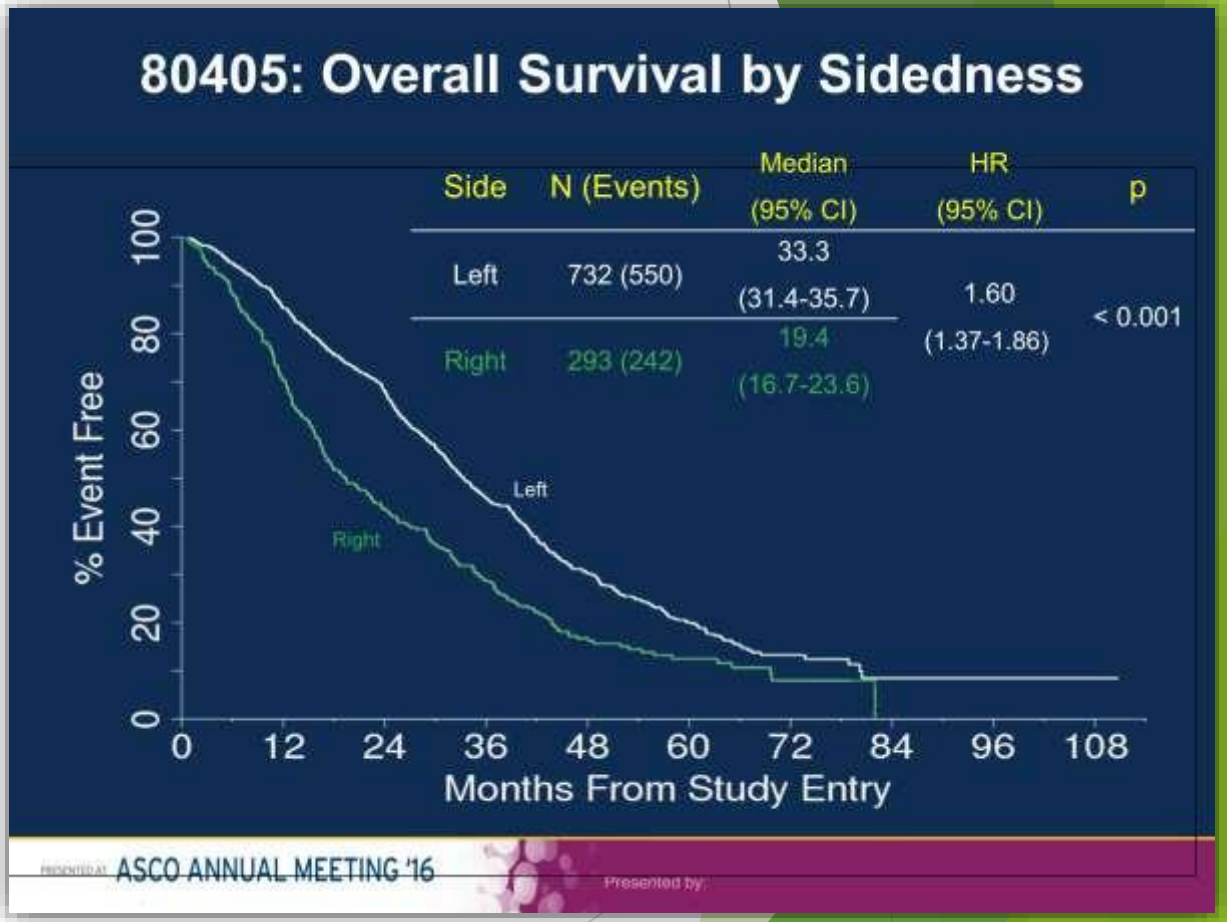
SWOG 80405 trial was a pivotal study.



Lenz et. al. ASCO 2016

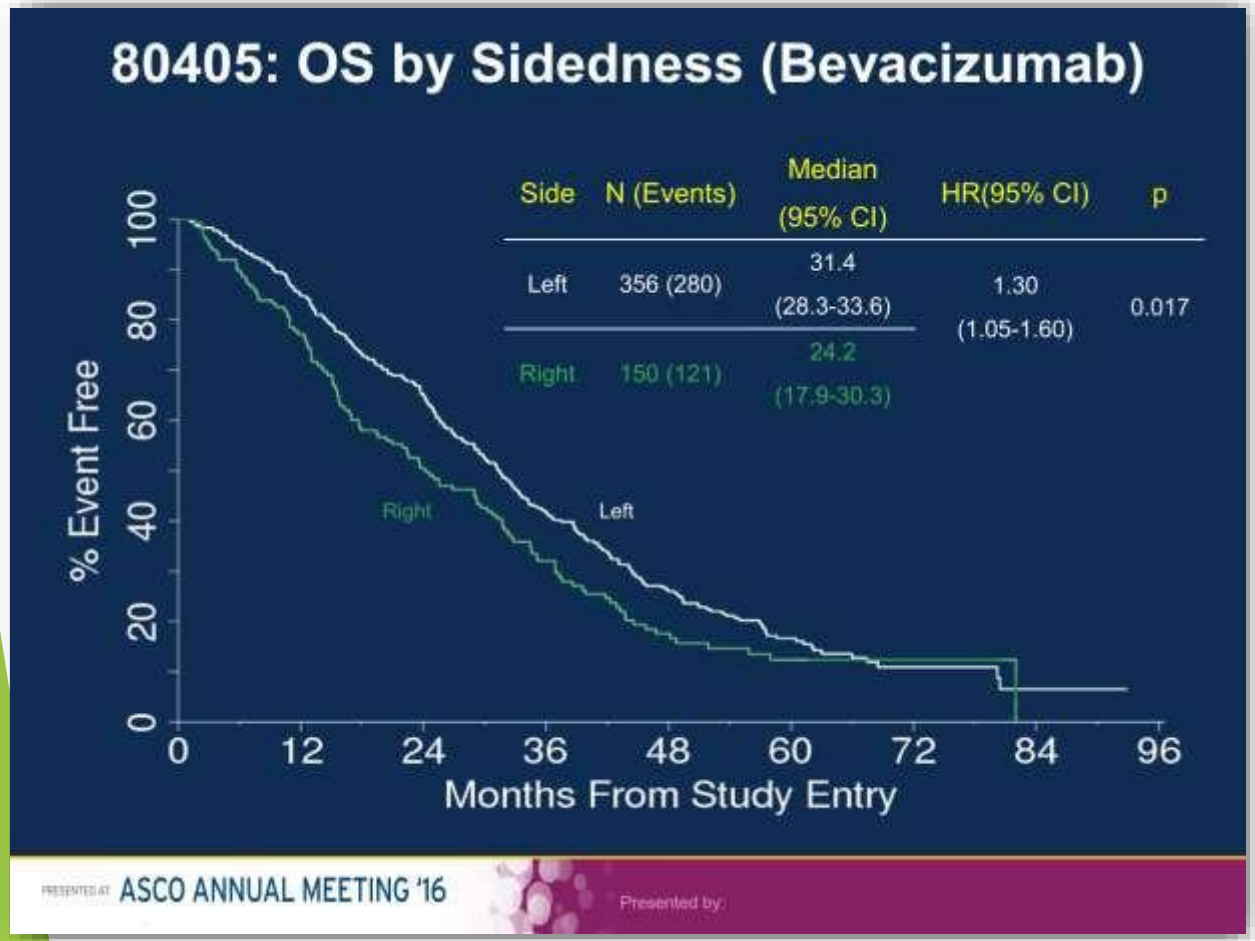
Left sided CRC do better overall.

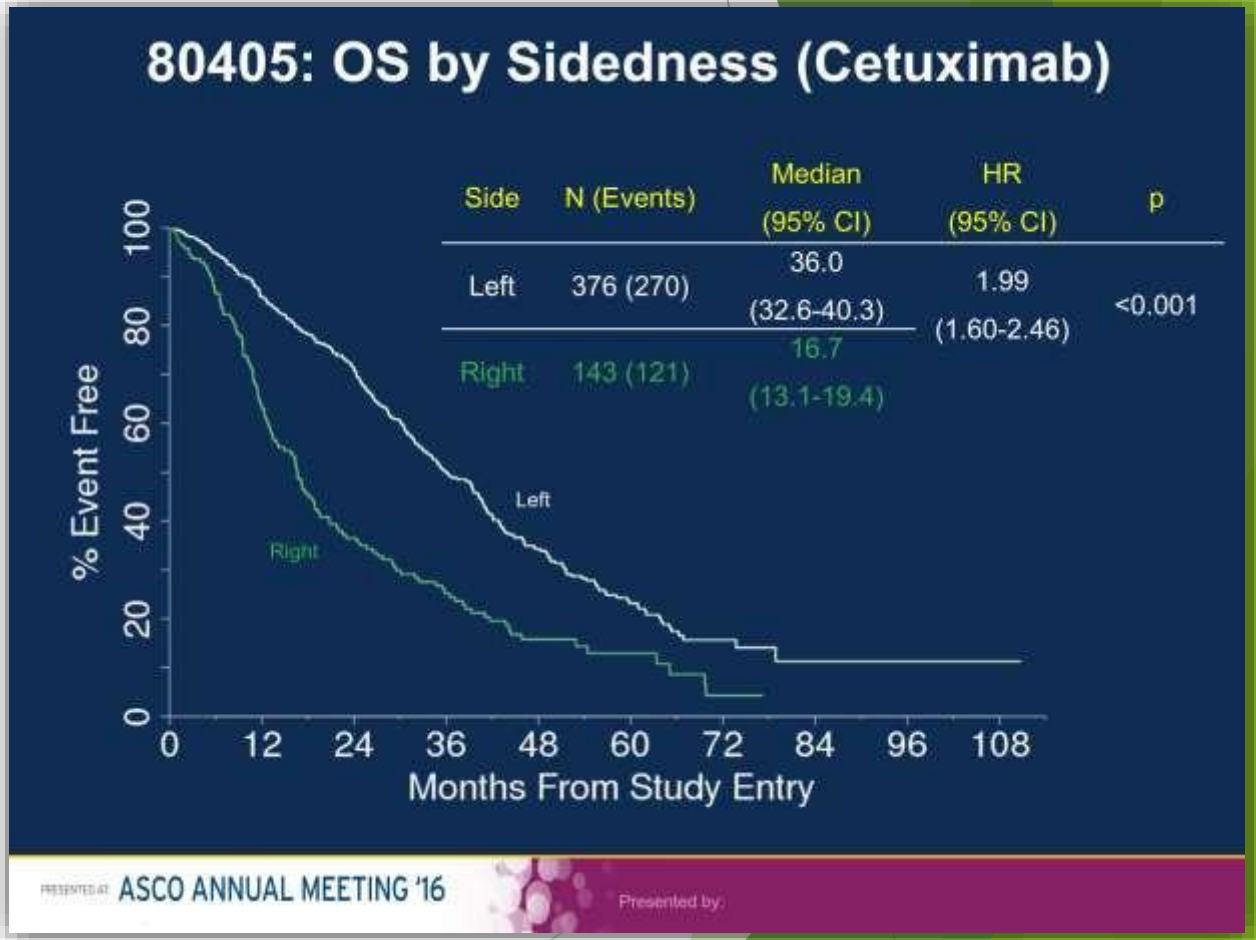




Lenz et. al. ASCO 2016

Left sided CRC do better on cetuximab.





Venook et. al. ASCO 2016

Colorectal Cancer Biomarkers in the Era of Personalized Medicine

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Abstract: The 5-year survival probability for patients with metastatic colorectal cancer has not drastically changed over the last several years, nor has the backbone chemotherapy in first-line disease. Nevertheless, newer targeted therapies and immunotherapies have been approved primarily in the refractory setting, which appears to benefit a small proportion of patients. Until recently, rat sarcoma (*RAS*) mutations remained the only genomic biomarker to assist with therapy selection in metastatic colorectal cancer. Next generation sequencing has unveiled many more potentially powerful predictive genomic markers of therapy response. Importantly, there are also clinical and physiologic predictive or prognostic biomarkers, such as tumor sidedness. Variations in germline pharmacogenomic biomarkers have demonstrated usefulness in determining response or risk of toxicity, which can be critical in defining dose intensity. This review outlines such biomarkers and summarizes their clinical implications on the treatment of colorectal cancer. It is critical that clinicians understand which biomarkers are clinically validated for use in practice and how to act on such test results.

Keywords: biomarker; prognostic; predictive; somatic; germline; pharmacogenomics; personalized medicine

Table 1. Side-specific features in colorectal cancer (CRC).

Location	Right-Side (Appendix to Hepatic Flexure)	Left-Side (Splenic Flexure to Rectum)
Embryonic Origin	Midgut	Hindgut
Demographics	Female Older	Male [a] Younger [a]
Prognosis	Worse prognosis in stage III + IV Worse response to EGFR inhibitors	Better survival in stage III + IV Better response to EGFR inhibitors
Cellular type	Goblet-like/mucinous Inflammatory CMS1 (MSI immune) CMS3 (metabolic) CCS2	Enterocyte Transit-amplifying CMS2 (canonical) CMS4 (mesenchymal) CCS1
Molecular Alterations	*BRAF* *dMMR* *EGFR*	*TP53* *APC* [a] *HER-2* [a]
Microbiology	Predominant biofilms Increased fusobacterium	Rare biofilms

Table 2. Summary of somatic biomarkers and clinical implications in colorectal cancer.

Biomarker	Predictive, Prognostic, or both?	Frequency of Mutation in mCRC	Clinical Implication	Guideline Recommendation available?
RAS	Both	40% *KRAS* 3–5% *NRAS* <1% *HRAS*	Presence of RAS wild-type tumors allows for use of anti-EGFR monoclonal antibodies. Possible worse prognosis for mCRC liver-resection.	NCCN recommends extended *RAS* (K-, N- and H-) genomic testing for prognosis and treatment options [28].
BRAF	Both	8–12%	Portends worse OS. Possibly less benefit when treated with anti-EGFR monoclonal antibodies. Combination BRAF inhibition has poor response rates.	Recommend genomic testing; no treatment recommendations or guidelines available [28].
MEK	Both	Unknown	MEK targeting may require triple inhibition of *RAS*, *BRAF*, and *MEK*.	No treatment recommendations or guidelines available
HER2	Predictive	3–4% mCRC 5%*RAS* WT mCRC	Progression of *RAS* wild-type tumor with HER2 amplification may benefit from dual HER2 inhibition.	No treatment recommendations or guidelines available

Table 2. *Cont.*

Biomarker	Predictive, Prognostic, or both?	Frequency of Mutation in mCRC	Clinical Implication	Guideline Recommendation available?
MSI/dMMR	Both	15% of stage II–III 4–5% of stage IV	Prognosis is stage-dependent: Better in stage II and potentially worse in more advanced stages. Presence of MSI predictive of nonresponse to 5-FU but not validated. dMMR may predict an increased response to an immune checkpoint.	NCCN recommends MSI testing for patients with mCRC. It is also recommended in patients with high-risk for Lynch Syndrome and/or for prognosis determination [28].
PIK3CA	Potentially predictive	10–20% of all CRC	Better outcomes with post-operative aspirin use. Studies show possible resistance to anti-EGFR therapy, but this could be exon-specific	No treatment recommendations or guidelines available
TILs	PrognosticPredictive value is debated	No consensus due to the variability of measurement methods	Most studies show improved prognosis in all stages of CRC however large more uniform studies are necessary for consensus	No treatment recommendations or guidelines available
POLE	Prognostic and predictive values being investigated	0.65–12.3%	Variability in outcomes based on current studies. More responsive to immunotherapy in other tumor types but currently under investigation in CRC.	No treatment recommendations or guidelines available

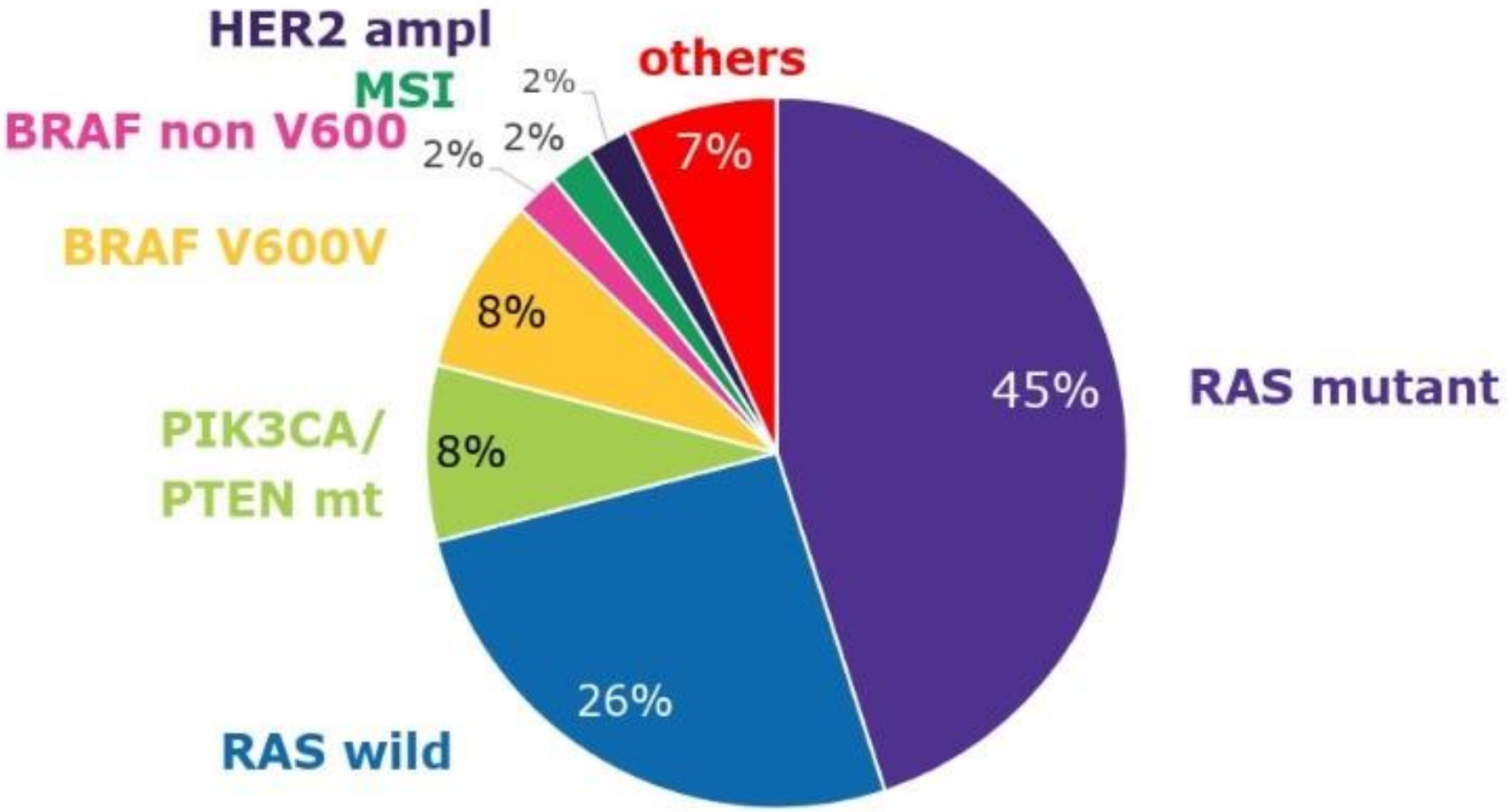
RAS—rat sarcoma virus; KRAS—Kirsten rat sarcoma virus; EGFR—epidermal growth factor receptor; BRAF—v-RAF murine sarcoma viral oncogene homolog B; mCRC—metastatic colorectal cancer; OS—overall survival; MEK—mitogen activated protein kinase; HER2—human epidermal growth factor receptor; WT—wild type; MSI—microsatellite instability; dMMR—deficient mismatch repair; PIK3CA—phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha; TIL—tumor infiltrating lymphocyte; POLE—polymerase epsilon, catalytic subunit.

Table 3. Summary of germline pharmacogenomic biomarkers and clinical implications in colorectal cancer.

Biomarker	Predictive, Prognostic, or both?	Frequency of Mutation [a]	Clinical Implication	Guideline Recommendation Available?
DPYD	Predictive	Heterozygous: 5–8% Homozygous variant: <0.1%	Polymorphisms resulting in reduced DPD enzyme function significantly increase the risk of fluoropyrimidine-related toxicities and possibly death	- Dosing recommendations available from CPIC [79] and DPWG [80] - Mentioned under "Warnings and Precautions" in FDA package insert [81]
UGT1A1	Predictive	Heterozygous (*1/*28): 40–45% Homozygous variant (*28/*28): 10–15%	Polymorphisms resulting in reduced UGT1A1 activity significantly increase the risk of irinotecan-related toxicities, particularly at higher doses. Wild type patients may tolerate higher than standard doses.	- Dosing recommendations available from DPWG [80] - Mentioned under "Warnings and Precautions" in FDA package insert [82]
TYMS	Predictive	3R/3R: 25% 2R/2R: 20%	Low *TYMS* expression (2R/2R) may increase 5-FU cytotoxicity, whereas high expression (3R/3R) may decrease 5-FU cytotoxicity.	No treatment recommendations or guidelines available
MTHFR	Predictive	C677T MAF: 30–40% A1298C MAF: 30–35%	Reduction in MTHFR activity increases 5,10-MTHF and increases 5-FU cytotoxicity.	No treatment recommendations or guidelines available
ERCC1/2	Both	rs11615C>T MAF: 45–50% rs13181T>G MAF: 30%	Mutations impairing the NER pathway can render cancers more sensitive to platinum treatment.	No treatment recommendations or guidelines available
VEGF	Both	rs833061 MAF: 35–40% rs699947 MAF: 32–40%	Presence of *VEGFA* SNPs, which reduce expression, may improve bevacizumab response and survival outcomes.	No treatment recommendations or guidelines available

[a] Allele or genotype frequencies are estimated for Caucasian or White populations. DPYD or DPD—dihydropyrimidine dehydrogenase; CPIC—Clinical Pharmacogenetics Implementation Consortium; DPWG—Dutch Pharmacogenetics Working Group; FDA—Food and Drug Administration; UGT1A1—uridine diphosphate glucuronosyltransferase 1A1; TYMS—thymidylate synthase; MTHFR—methylenetetrahydrofolate reductase; ERCC—excision repair complementing complex; NER—nucleotide excision repair; VEGF—vascular endothelial growth factor; MAF—minor allele frequency.

Genomic markers in CRC



Slide courtesy of R. Dienstmann

What biomarkers do I need to consider when treating a mCRC patient?

Recommended for routine assessment (ESMO and NCCN)

RAS
BRAF
MSI

An option for selected patients (ESMO)

UGT1 A1
DPD

Assessment not recommended (ESMO and/or NCCN)

TGF-α/β
EGFR
MET
ERCC 1
HER3
PIK3 CA
PTEN
AREG / EREG
TS/ TSER

Assessment not recommended but emerging data

HER2
miR

MSI, microsatellite instability; DPD, dihydropyrimidine dehydrogenase;
UGT1A1, uridine diphosphate glucuronosyltransferase 1 family, polypeptide A1;
TS/TSER, thymidylate synthase/thymidylate synthase enhancer region;
PIK3CA, phosphatidylinositol 4,5-bisphosphate 3-kinase catalytic subunit alpha isoform;
PTEN, phosphatase and tensin homolog; AREG/EREG, amphiregulin/eregulin;
TGFα/β, transforming growth factor α/β; EGFR, epidermal growth factor receptor;
MET, mesenchymal-epidermal transition; ERCC1, excision repair cross-complementation group 1;
HER3, human epidermal growth factor receptor 3; miR, microRNA

NCCN Colon Cancer Guidelines Version 1.2019;

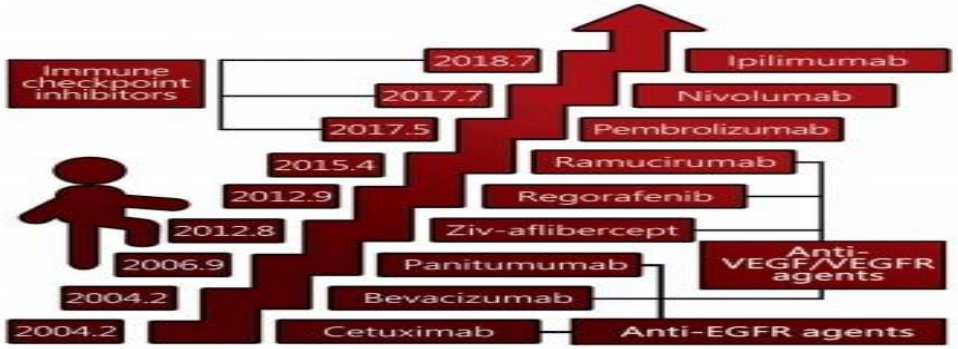
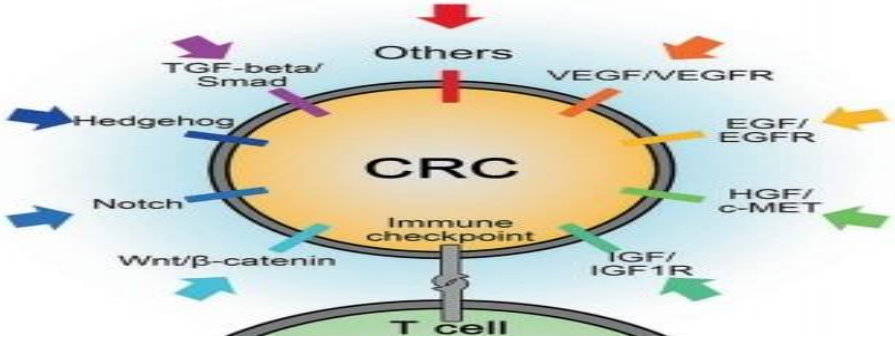


Fig. 1 United States of America Food and Drug Administration (FDA)-approved targeted agents in colorectal cancer. VEGF: vascular endothelial growth factor; VEGFR: vascular endothelial growth factor receptor; EGFR: epidermal growth factor receptor



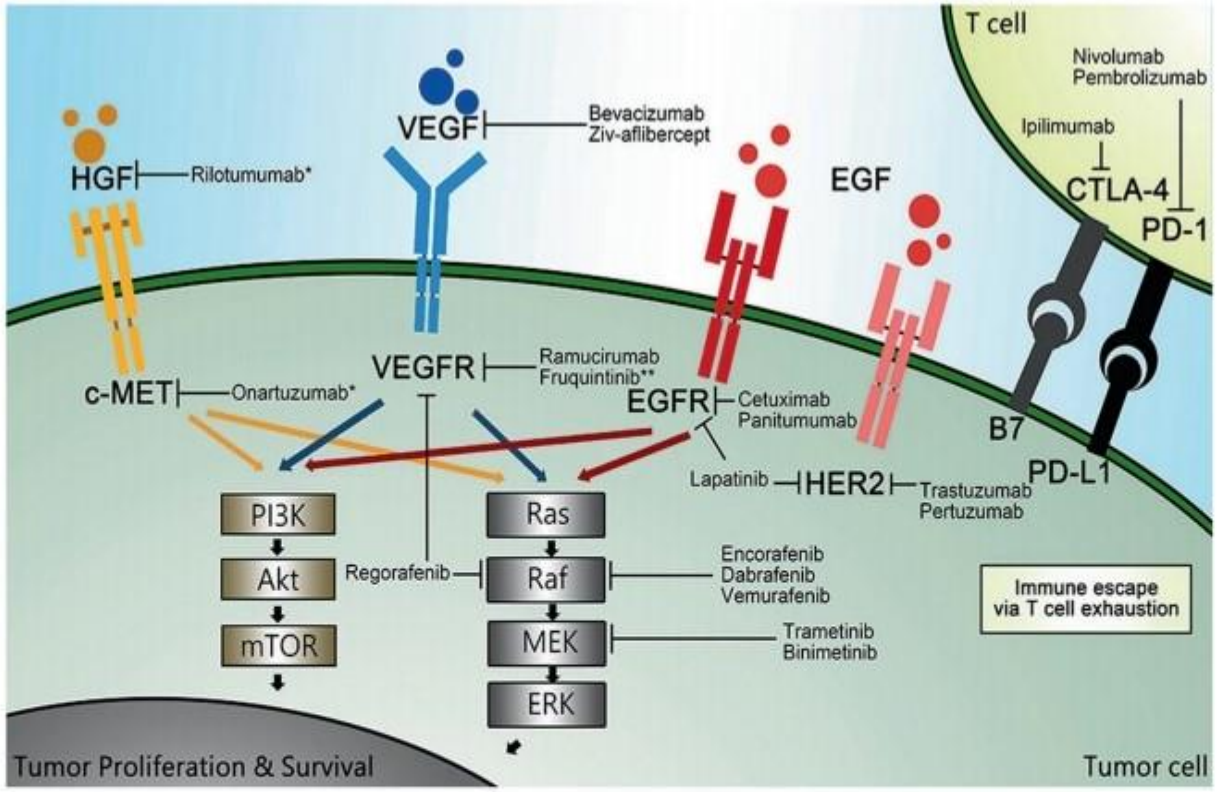
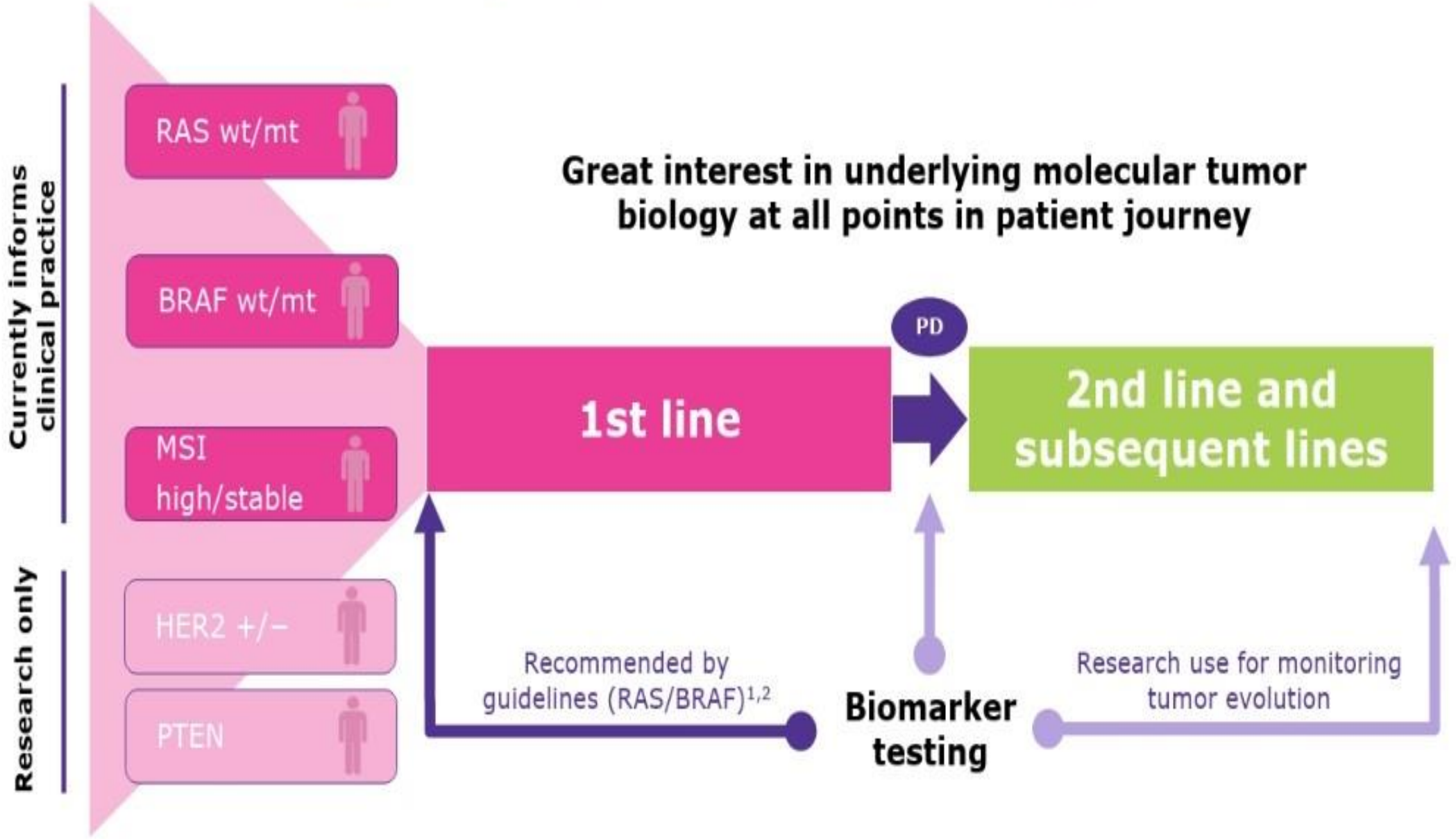
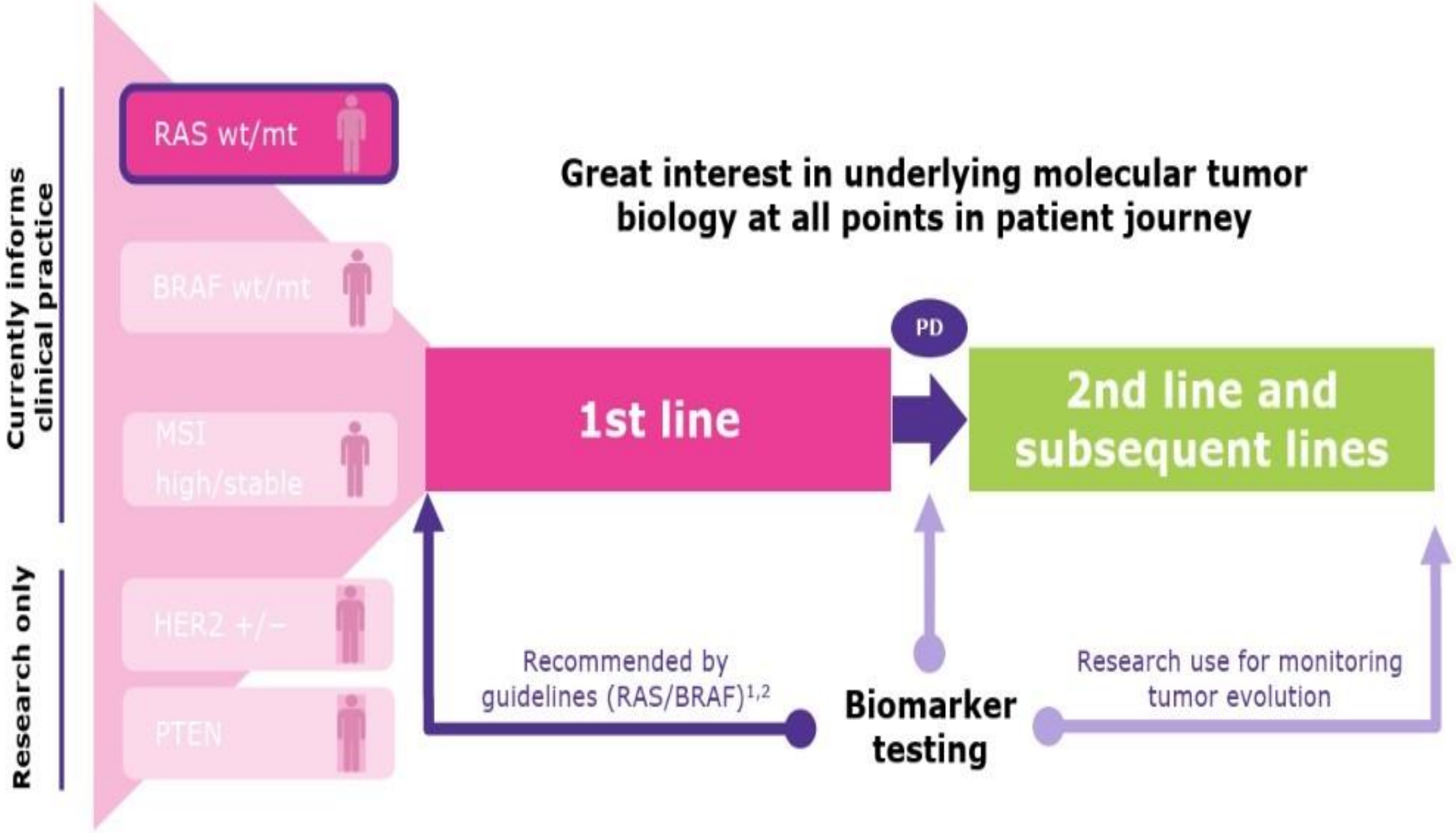


Fig. 3 Overview of National Comprehensive Cancer Network (NCCN)-recommended targeted agents. HGF: hepatocyte growth factor; c-MET: mesenchymal–epithelial transition factor; VEGF: vascular endothelial growth factor; VEGFR: vascular endothelial growth factor receptor; EGFR: epidermal growth factor receptor; EGF: epidermal growth factor; HER2: human epidermal growth factor 2; CTLA-4: cytotoxic T lymphocyte-associated antigen 4; PD-1: programmed death-1; PD-L1: programmed death ligand 1; PI3K: phosphoinositide 3-kinase; AKT: protein kinase B, also known as PKB; mTOR: mammalian target of rapamycin; MEK: mitogen-activated protein kinase; ERK: extracellular signal-regulated kinase. *These agents have not been recommended by the NCCN. **This agent has been approved by the National Medical Products Administration of China (NMPA), but not by the United States of America Food and Drug Administration (FDA)

Biomarkers are gaining importance in the management of mCRC

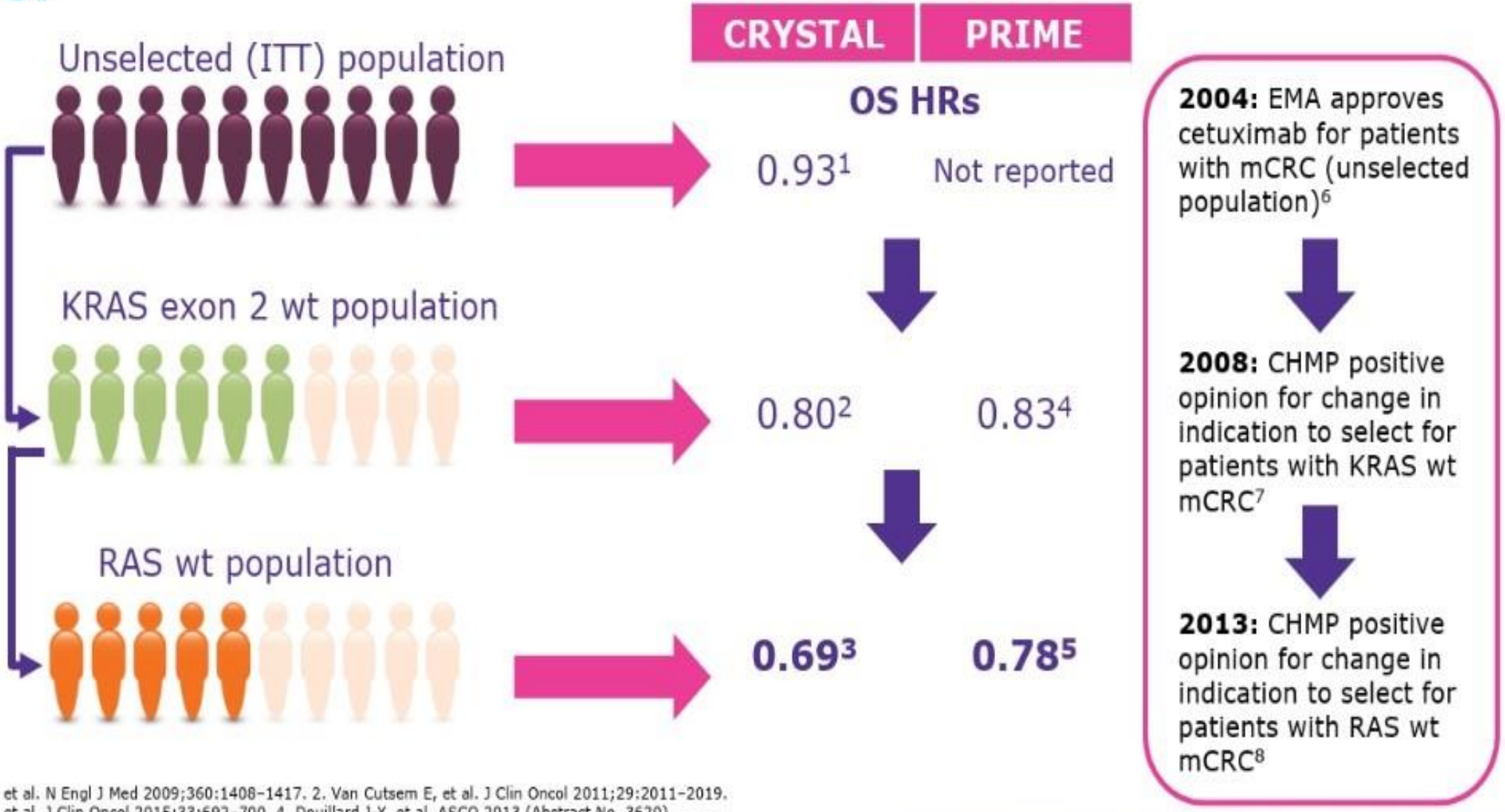


Biomarkers are gaining importance in the management of mCRC

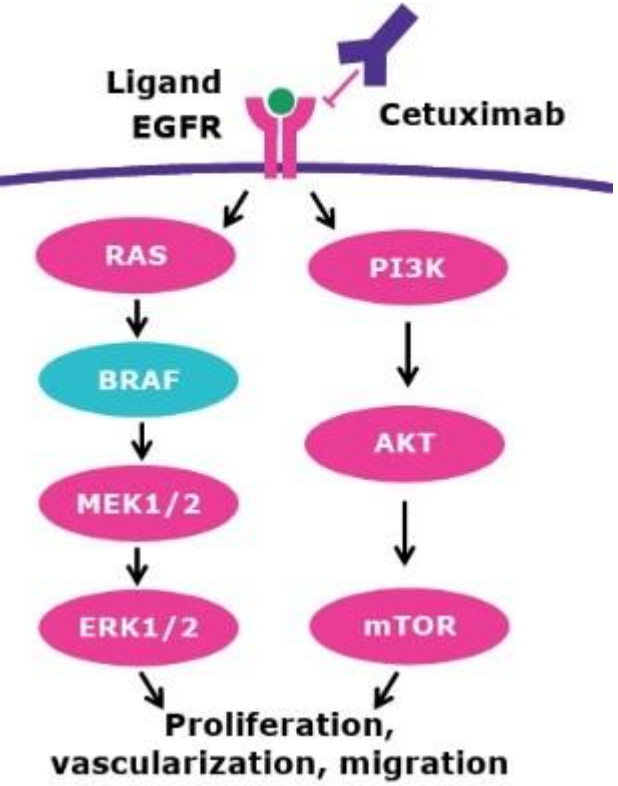


The RAS family of signaling proteins

RAS wt patient selection increases OS benefit of anti-EGFR addition to CT



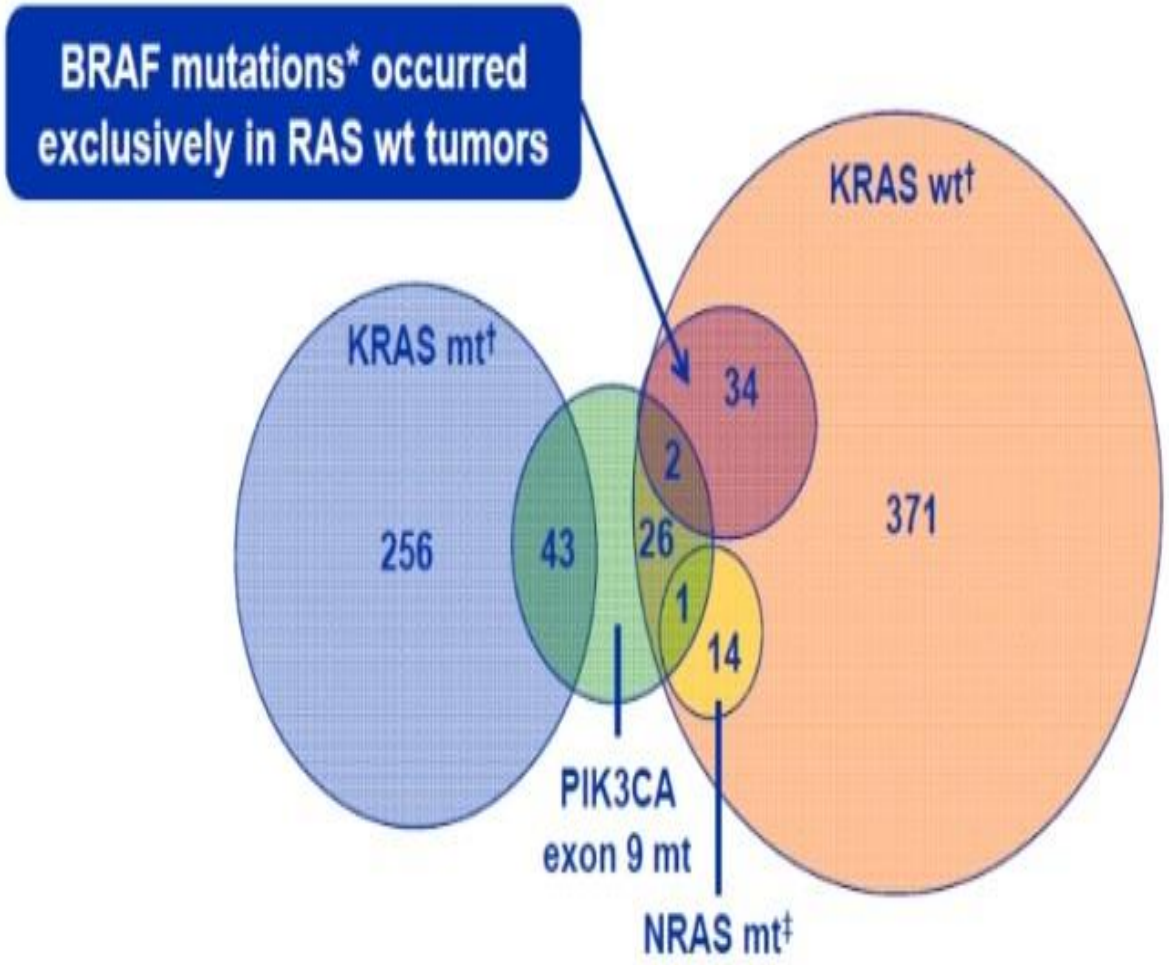
1. Van Cutsem E, et al. N Engl J Med 2009;360:1408–1417. 2. Van Cutsem E, et al. J Clin Oncol 2011;29:2011–2019.
3. Van Cutsem E, et al. J Clin Oncol 2015;33:692–700. 4. Douillard J-Y, et al. ASCO 2013 (Abstract No. 3620).
5. Douillard J-Y, et al. N Engl J Med 2013;369:1023–1034. 6. EMA Erbitux EPAR Scientific Discussion http://www.ema.europa.eu/docs/en_GB/document_library/EPAR_-_Scientific_Discussion_-_Variation/human/000558/WC500029116.pdf.
7. EMA CHMP positive opinion for Erbitux summary 2008. http://www.ema.europa.eu/docs/en_GB/document_library/Summary_of_opinion/human/000558/WC500059827.pdf. 8. EMA CHMP positive opinion for Erbitux summary 2013. http://www.ema.europa.eu/docs/en_GB/document_library/Summary_of_opinion/human/000558/WC500155463.pdf. (All accessed 20 February 2017).



Distribution of BRAF mutation vs other mutations

Retrospective consortium analysis of tumor samples from 747 patients with chemorefractory mCRC treated with Erbitux + CT[1]

- Occurs in 10%–15% of patients and confers a poor prognosis[1-3]



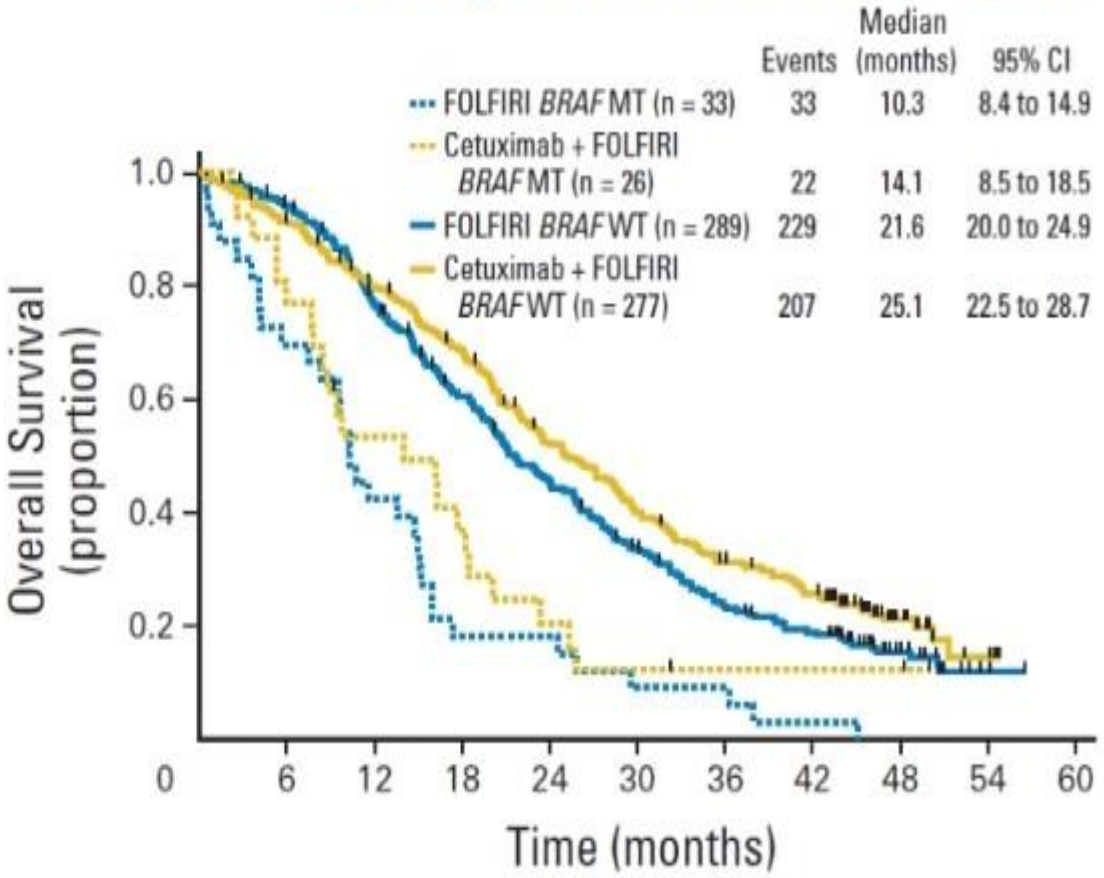
BRAF mt* is a marker for poor prognosis but does it have a predictive role for efficacy of anti-EGFR therapy?

Many studies have considered BRAF mt but were not powered to determine efficacy

e.g. CRYSTAL and OPUS studies[1]

Subanalysis of Crystal Trial:

according to BRAF V600E mutation status



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Many studies have considered BRAF mt but were not powered to determine efficacy

e.g. CRYSTAL and OPUS studies[1]

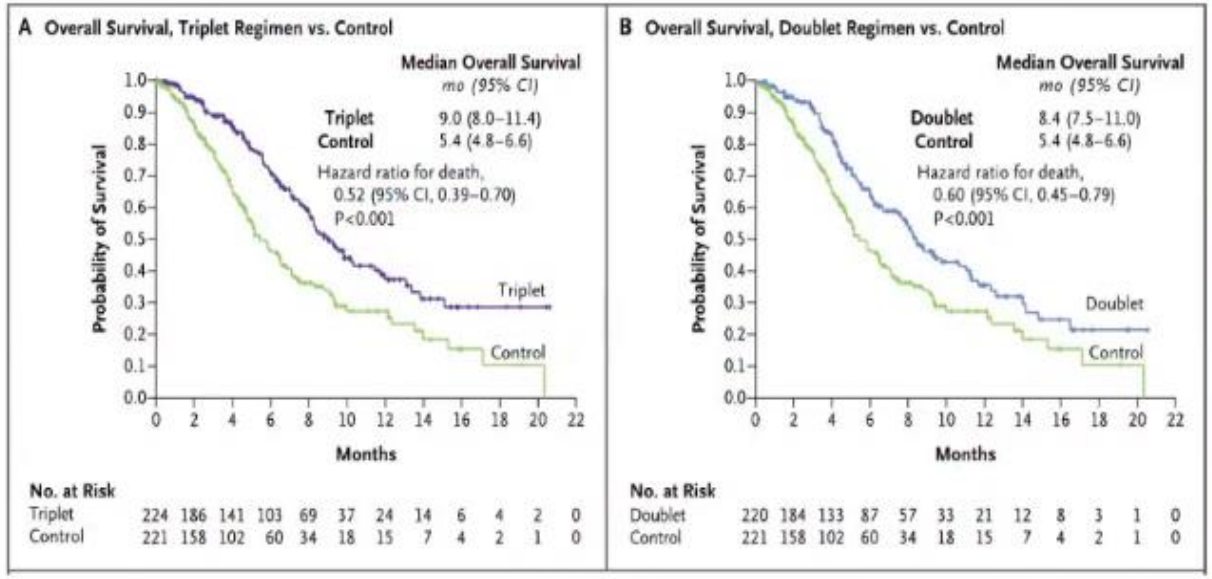
Pietrantonio[2] and Rowland[3] conducted meta-analyses to assess efficacy of anti-EGFR agents in patients with BRAF mt

Overall survival

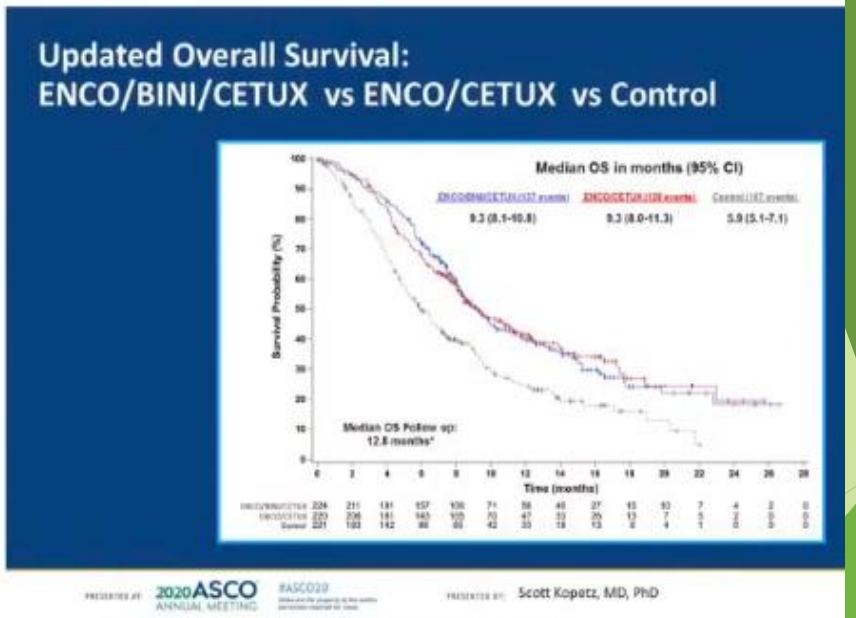
	Anti-EGFR therapy ± CT vs SOC HR (95% CI)	p value
Pietrantonio *et al.*[2]	No test for interaction	
(K)RAS wt /BRAF mt	0.91 (0.62–1.34)	**0.63**
Rowland *et al.*[3]		
(K)RAS wt /BRAF wt n=2748	0.81 (0.70–0.95)	0.009
	Test for interaction: p=0.43 (not significant)	
(K)RAS wt /BRAF mt n=351	0.97 (0.67–1.41)	0.88

Cetuximab, Encorafenib (anti-V600E) alone / with Binimetinib (anti-MEK)

BEACON trial, BRAF mut, 2nd/3rd line



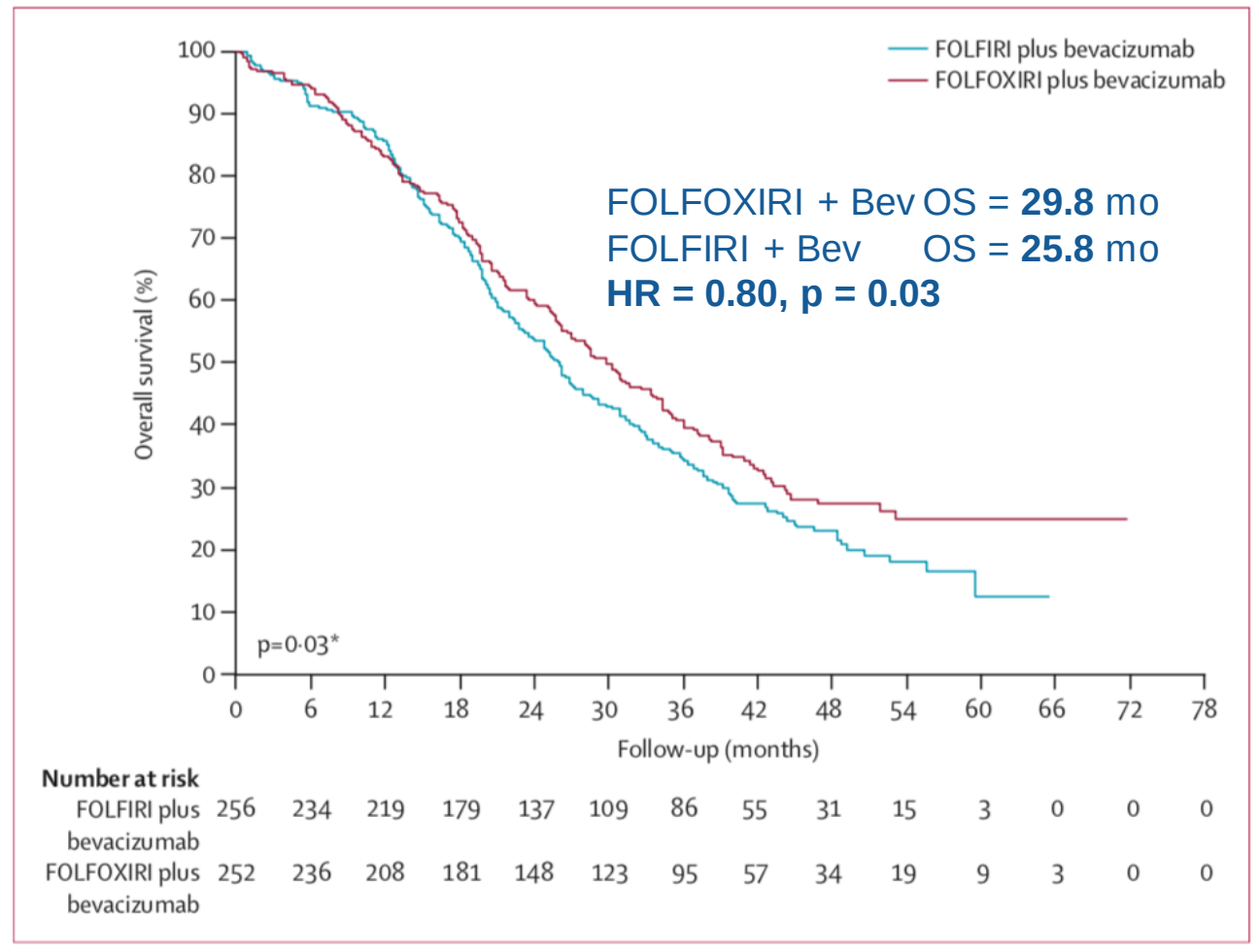
Kopetz et al., New Engl J Med 2019



Kopetz et al., ASCO 2020

TRIBE Study: FOLFIRI+ Bev vs FOLFOXIRI + Bev

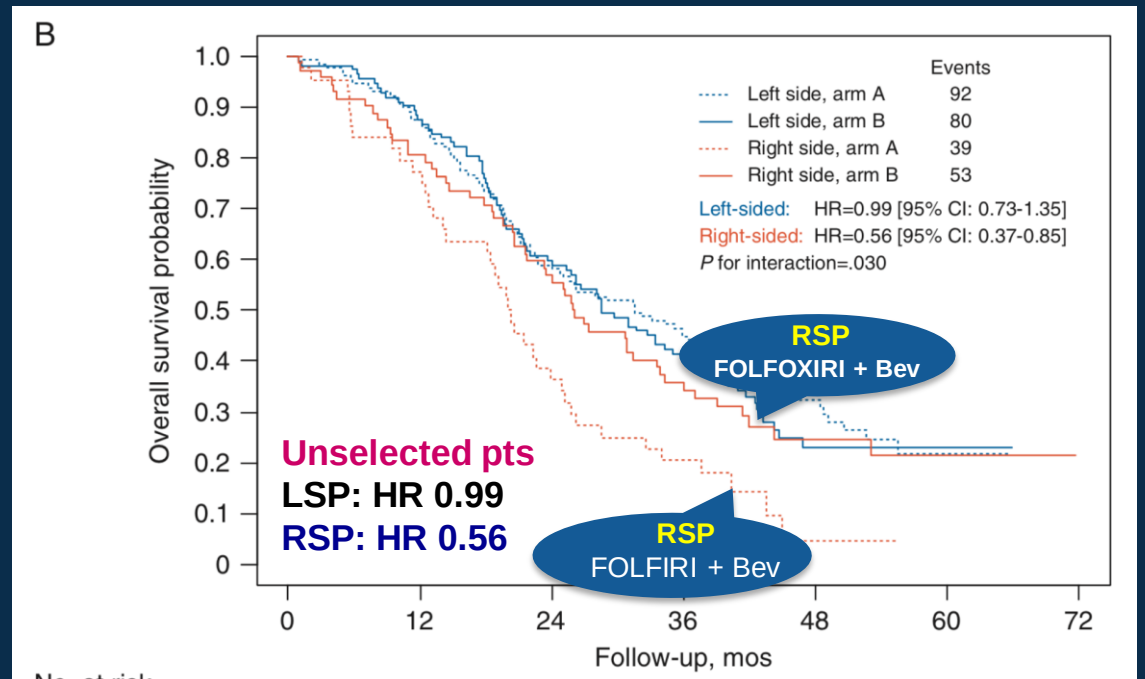
	FOLFIRI + Bev	FOLFOXIRI + Bev
ORR	54%	65%
PFS	9.7 mo	12.3 mo
Neutropenia	20.5%	50%
Febrile neutropenia	6.3%	8.8%
Diarrhea	10.6%	18.8%



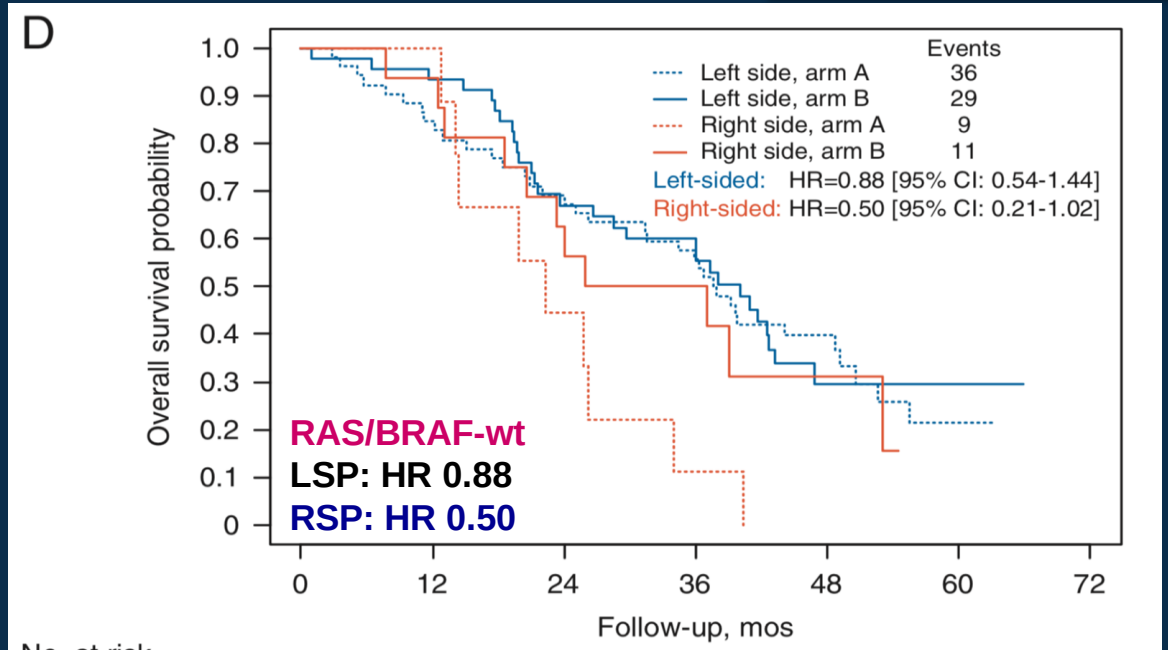
Cremolini et al. Lancet Oncol 2015

TRIBE: Impact of Sidedness on OS

FOLFIRI + Bev
FOLFOXIRI + Bev



No. at risk							
Left side, arm A	129	113	75	57	23	2	0
Left side, arm B	113	98	65	43	13	4	0
Right side, arm A	44	34	16	9	1	0	0
Right side, arm B	72	58	41	23	9	2	0



No. at risk							
Left side, arm A	52	44	35	29	12	1	0
Left side, arm B	47	43	29	25	7	3	0
Right side, arm A	9	9	4	1	0	0	0
Right side, arm B	16	15	10	6	2	0	0

RSP: FOLFOXIRI plus bevacizumab may be regarded as a preferred option in pts fit for combination independent of their molecular status (*addition of oxaliplatin to FOLFIRI beneficial*)

LSP: Doublet plus bevacizumab remains the prefered option

ol 2018

TRIBE: Impact of Sidedness on OS

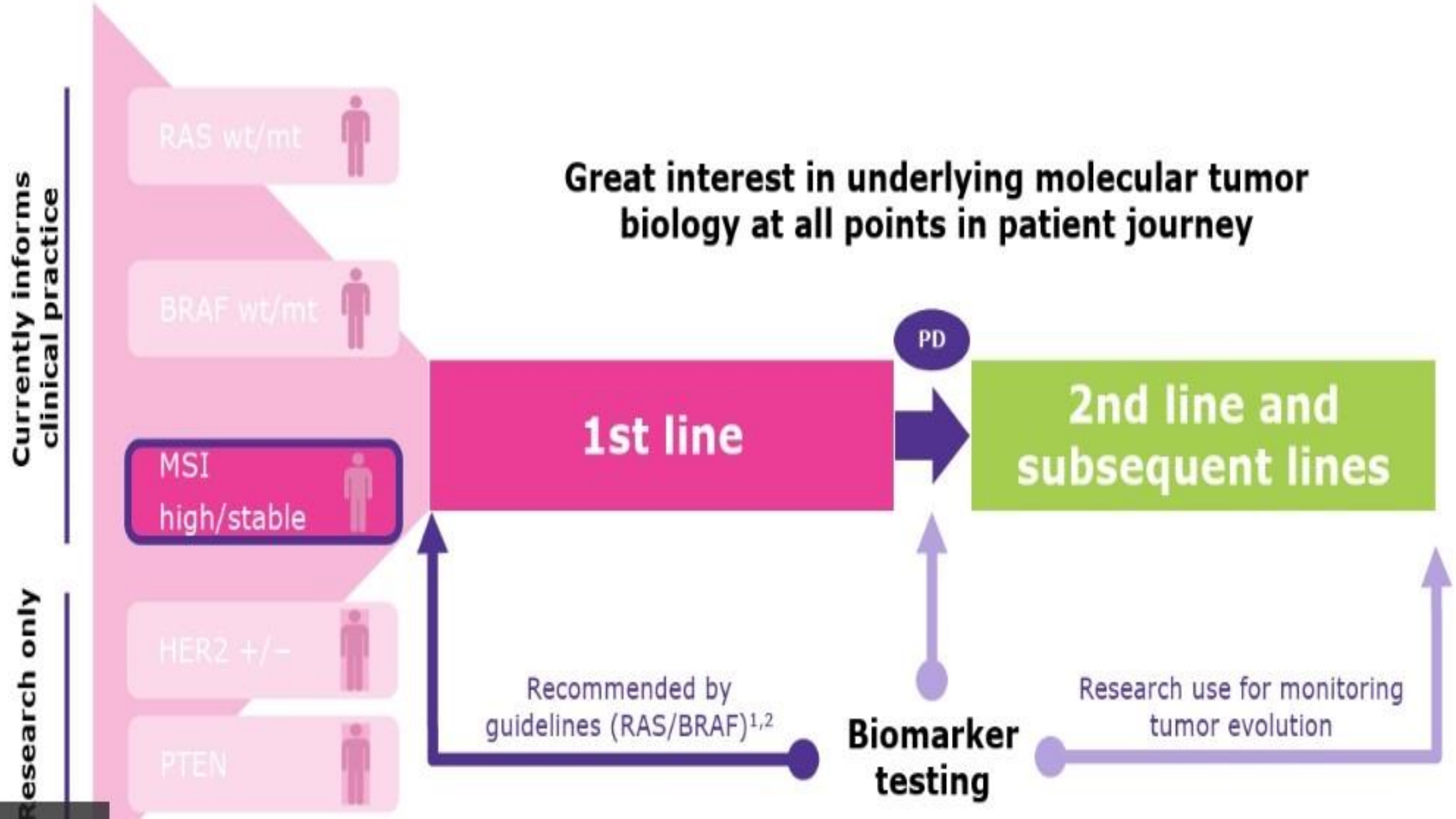
Unplanned subgroup analysis

	ITT Population				RAS wt				BRAF mt			
	Right Side		Left Side		Right Side		Left Side		Right Side		Left Side	
	FOLFOXIRI +bev *N*=72	FOLFIRI +bev *N*=44	FOLFOXIRI +bev *N*=113	FOLFIRI +bev *N*=129	FOLFOXIRI +bev *N*=16	FOLFIRI +bev *N*=9	FOLFOXIRI +bev *N*=47	FOLFIRI +bev *N*=52	FOLFOXIRI +bev *N*=8	FOLFIRI +bev *N*=7	FOLFOXIRI +bev *N*=6	FOLFIRI +bev *N*=4
PFS												
Median (mos)	11.2	9.4	10.7	11.0	13.4	10.1	14.1	11.5	9.0	4.2	6.5	10.9
HR [95% CI]	0.59 [0.40–0.88]		0.89 [0.68–1.16]		0.54 [0.24–1.22]		0.85 [0.56–1.29]		0.33 [0.12–0.92]		1.74 [0.48–6.30]	
P	0.099[a]				0.292[a]							
OS												
Median (mos)	26.0	20.2	28.6	31.6	31.5	22.3	40.0	37.7	20.4	9.4	12.0	25.2
HR [95% CI]	0.56 [0.37–0.85]		0.99 [0.73–1.35]		0.50 [0.21–1.02]		0.88 [0.54–1.44]		0.22 [0.07–0.67]		2.02 [0.50–8.15]	
P	0.030[a]				0.165[a]							
ORR												
Rate, %	63.9	54.6	64.6	56.6	81.3	66.7	63.8	65.4	37.5	42.9	66.7	50.0
OR [95% CI]	1.48 [0.68–3.26]		1.43 [0.84–2.44]		2.17 [0.33–14.06]		0.93 [0.41–2.13]		0.80 [0.10–6.35]		2.00 [0.15–26.73]	
P	0.942[a]				0.584[a]							

FOLFOXIRI plus bevacizumab may be regarded as a preferred first-line treatment option for clinically selected patients with right-sided mCRC irrespective of their RAS and BRAF mutational status

Cremolini et al. Lancet Oncol 2015

Biomarkers are gaining importance in the management of mCRC



MSI-H/dMMR in mCRC

A biomarker for therapeutic effect of immunotherapy

Approximately
4%
of mCRC tumors

- MMR genes are found in Lynch syndrome and occasionally in sporadic colon cancers.
- Their characteristic genetic signature of tumors is a high number of DNA replication errors.
- Tumors that lack the mismatch repair mechanism harbor are often hyper-mutated than tumors of the same type without such MMR defects.

MSI-H/dMMR in mCRC

A biomarker for therapeutic effect of immunotherapy

Approximately
4%
of mCRC tumors

MSI-H/dMMR associated with

Tumor neoantigens

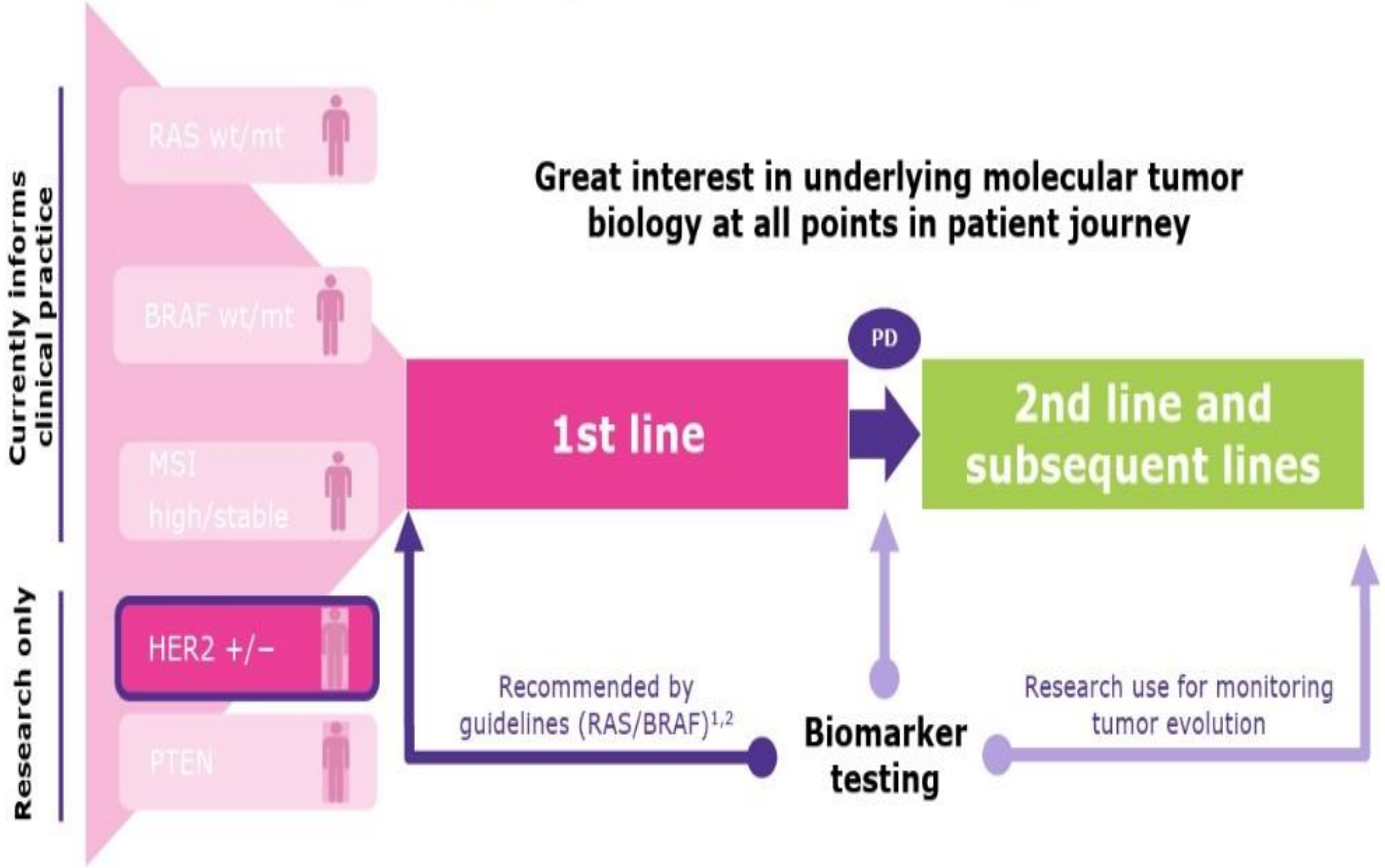
Immune cell checkpoint regulator expression

Tumor-infiltrating lymphocytes

New approvals and additions to guidelines

Immune checkpoint inhibitor	Population	Results
Pembrolizumab (anti-PD-1)	**dMMR/MSI-H** • Solid tumors; n=149 • **CRC (after PD on fluoropyrimidine, oxaliplatin, and irinotecan); n=90**[1]	• ORR (solid tumors): 39.6% • **ORR (mCRC): 36%**
	dMMR (n=11) vs pMMR (n=21) CRC[2]	• **ORR: 40%** (dMMR) **vs 0%** (pMMR) • Adjusted† OS HR: 0.18, p=0.05
Nivolumab (anti-PD-1)	**dMMR/MSI-H mCRC, ≥2 prior lines of therapy (n=74)**[3]	• **ORR: 27.0%** • DCR for ≥12 weeks: 62.2% • 12-month PFS rate: 45.6% • 12-month OS rate: 73.8%

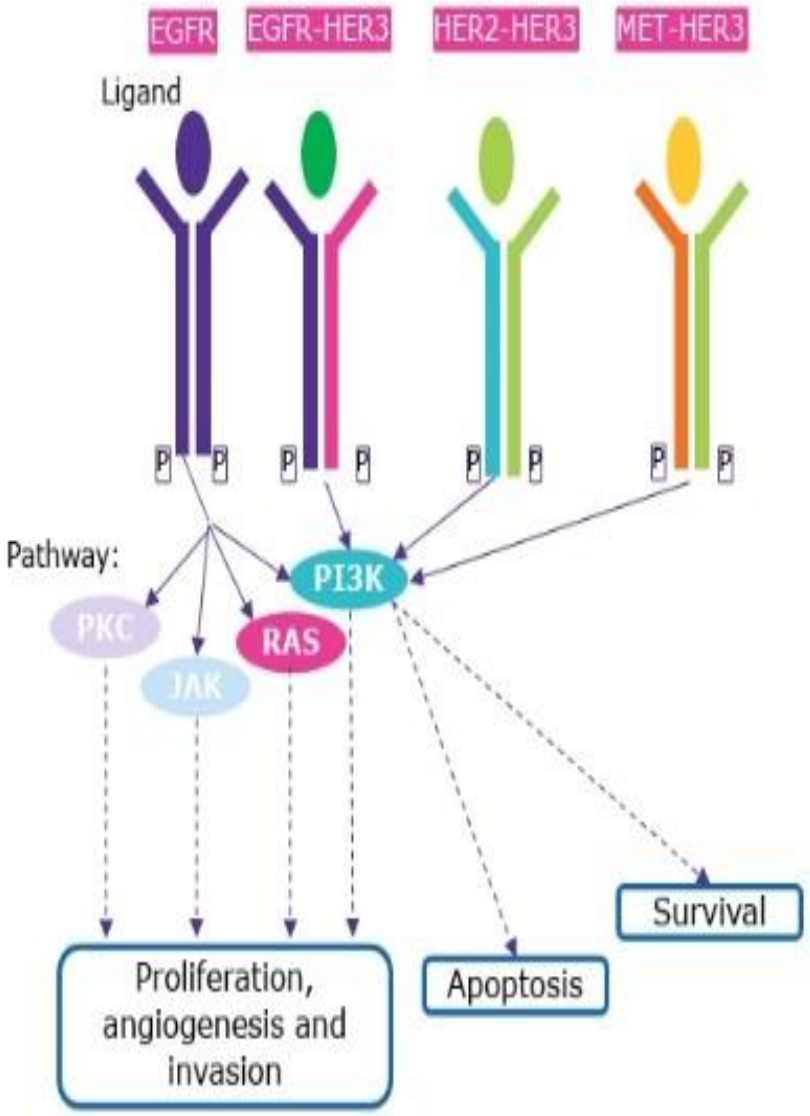
Biomarkers are gaining importance in the management of mCRC



HER family of proteins

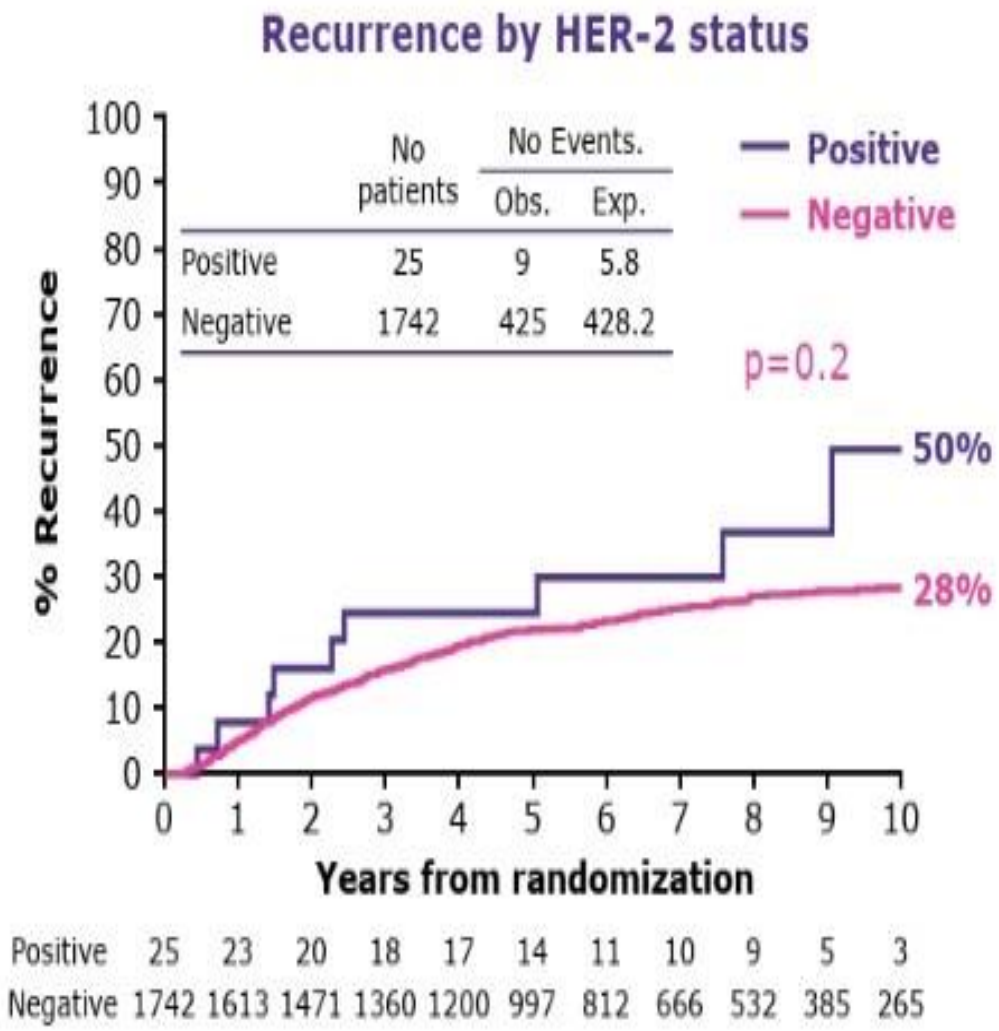
HER2: Structure and function

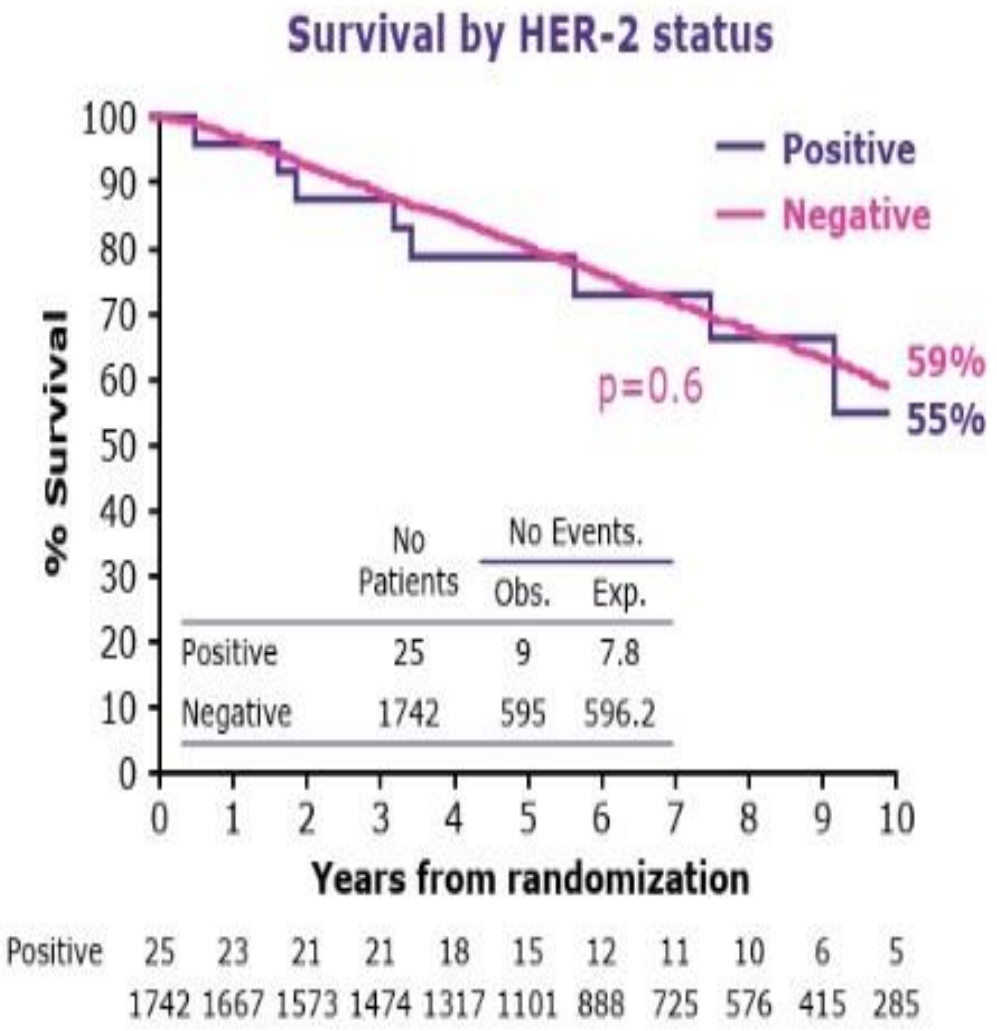
- HER2 is a member of the HER family of four receptor tyrosine kinases that includes EGFR (HER1)[1]
- Ligand binding to HER induces homo- or heterodimerization, activating MAPK, PI3K/AKT and PKC[2]
- HER2 has no direct ligand and may be physiologically constitutively active
  - It may also become active on heterodimerization with other HER proteins e.g. HER3 (activated by heregulin)[2]
- HER2 amplification/overexpression can increase the number of dimerization events, leading to downstream signaling pathway overactivation
  - This may promote oncogenesis and metastasis through prolonged signaling and is linked to the pathogenesis and progression of several human cancers, including CRC[1,2]



Is HER2 overexpression prognostic in stage II resected CRC (QUASAR*)?

No significant correlations between membranous HER2 overexpression and recurrence or OS in patients with stage II resected CRC[1]





Is HER2 overexpression prognostic in patients with mCRC (FOCUS and PICCOLO)?*

No significant association between HER2 and OS or PFS in mCRC[1]

		HER2 status (+ vs -), n	HR (95% CI)	p-value
OS	FOCUS	18 vs 826	0.81 (0.50–1.32)	NR
	PICCOLO	11 vs 434	0.98 (0.54–1.78)	NR
	COMBINED	29 vs 1260	0.87 (0.60–1.27)	0.48
	COMBINED (KRAS wt† only)	24 vs 442†	0.99 (0.65–1.51)	0.95
PFS	FOCUS	18 vs 826	0.68 (0.42–1.12)	NR
	PICCOLO	11 vs 434	0.80 (0.44–1.46)	NR
	COMBINED	29 vs 1260	0.73 (0.50–1.07)	0.11
	COMBINED (KRAS wt† only)	24 vs 442†	0.83 (0.54–1.28)	0.40

The number of patients with HER2 overexpression was small (29/1342; 2.2%)

HER2 overexpression/amplification in mCRC

Prognostic and/or predictive?

~5% of RAS wt/BRAF wt mCRC tumors[1]

No prognostic role

No significant association between HER2 overexpression and OS or PFS in mCRC[2*]

FOCUS/PICCOLO pooled analysis			
	HER2 status + vs - (n=260)	HR (95% CI)	p-value
OS	29 vs 1260	**0.87 (0.60–1.27)**	**0.48**
PFS	29 vs 1260	**0.73 (0.50–1.07)**	**0.11**

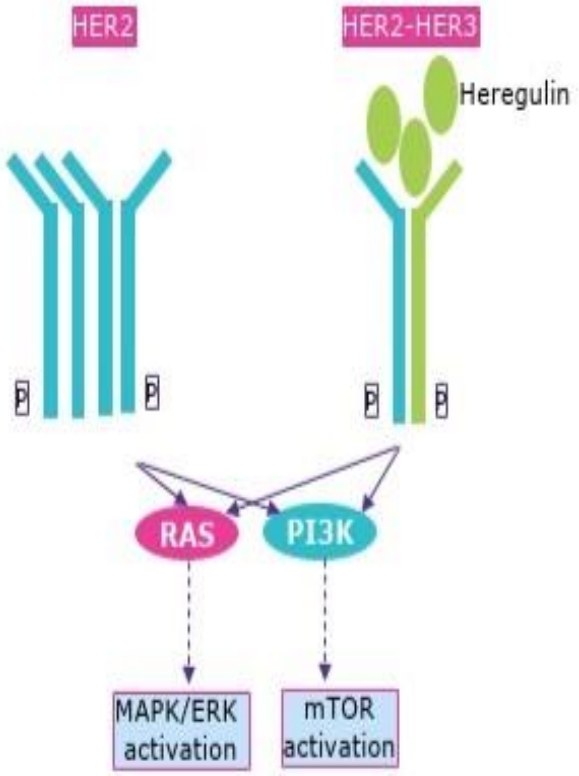
Potential predictive role

HER2 amplification associated with shorter PFS in patients who received anti-EGFR therapy in 2nd/3rd line[3*]

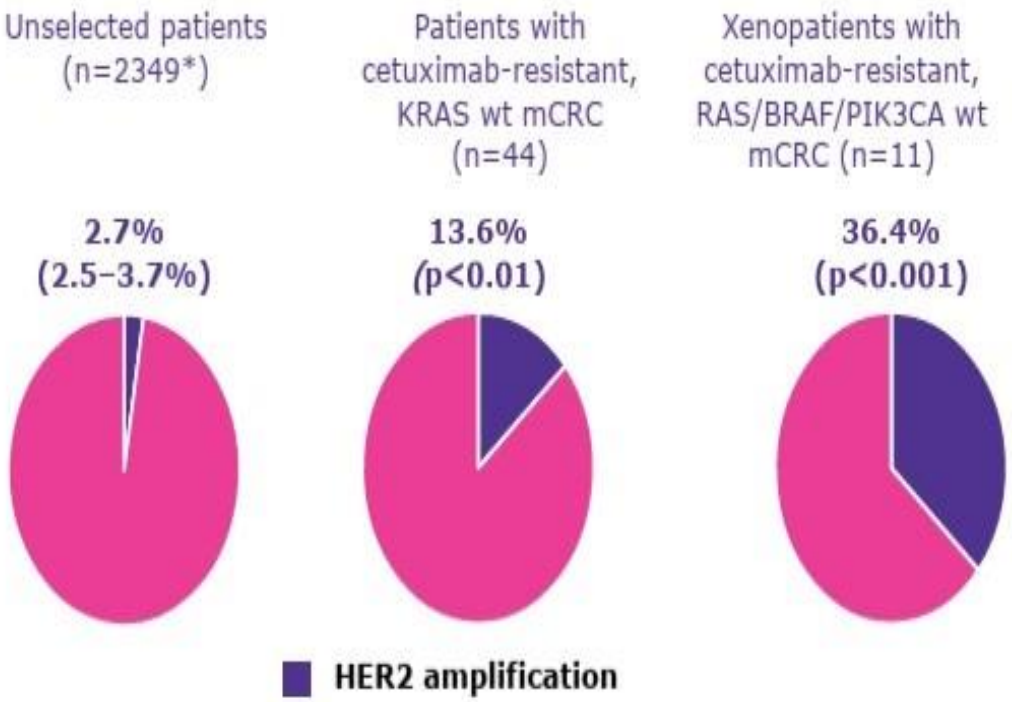
RAS wt/ BRAF wt patients	Therapy	Median PFS, months		
		HER2 amp+	HER2 amp-	p-value
Test cohort 1 (n=97)	1st line, non-anti-EGFR	9.7	10.1	0.848
	2nd/3rd line, anti-EGFR	**2.9**	**8.1**	**<0.001**
Validation cohort 2 (n=99)	1st line, non-anti-EGFR	13.7	11.3	<0.616
	2nd/3rd line, anti-EGFR	**2.9**	**9.3**	**<0.001**

HER2 activation may contribute to anti-EGFR resistance

Aberrant HER2 activation may bypass EGFR inhibition through activation of MAPK signaling independent of EGFR[1]



HER2 amplification in cetuximab-resistant patients with KRAS wt mCRC and RAS/BRAF/PIK3CA wt xenopatients[†2]



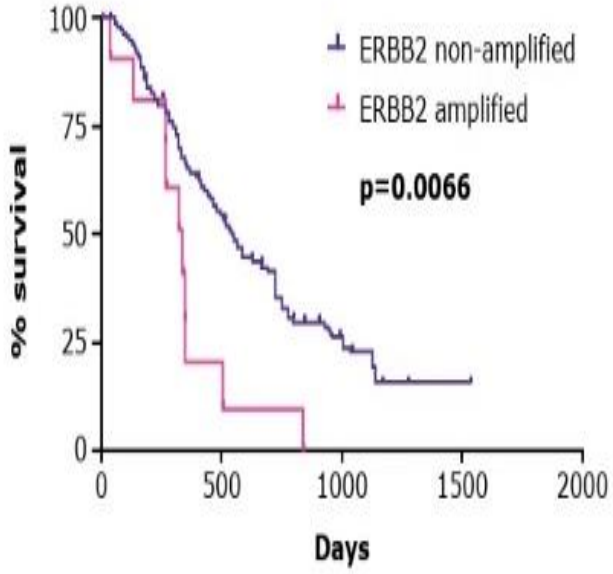
There was no evidence of HER2 amplification in any of 45 patient cases of KRAS wt mCRC that responded to anti-EGFR therapy[2]

HER2 activation may contribute to anti-EGFR resistance in mCRC

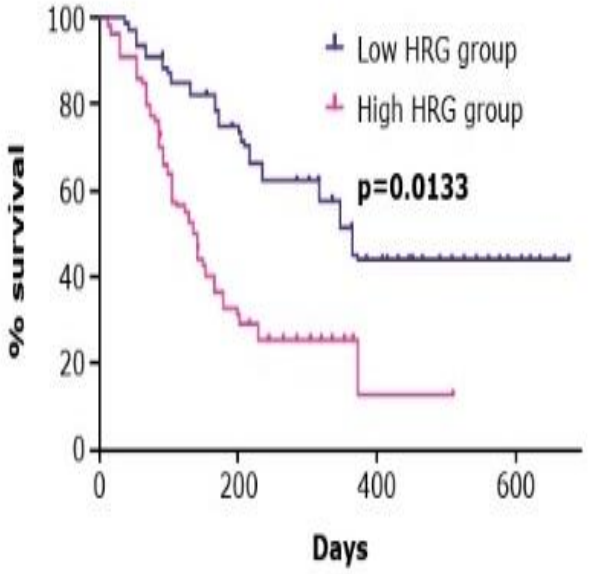
HER2 amplification (+) vs (-) and high vs low plasma heregulin associated with shorter OS in patients treated with cetuximab ± CT (KRAS wt)

De novo resistance to anti-EGFR:*

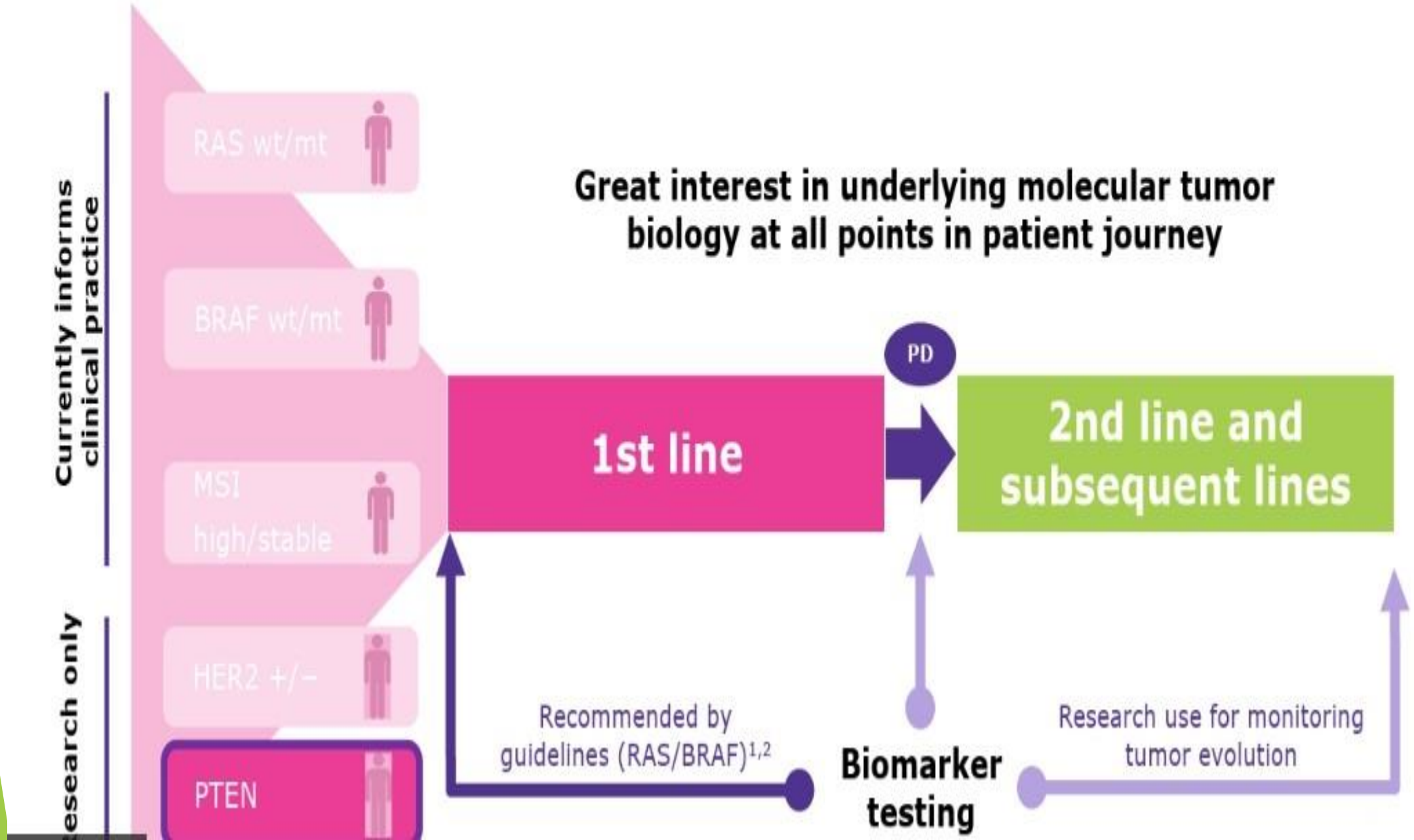
OS: HER2 amplified (n=11) vs non-amplified (n=171)



OS: Plasma heregulin: high (n=24) vs low (n=18)

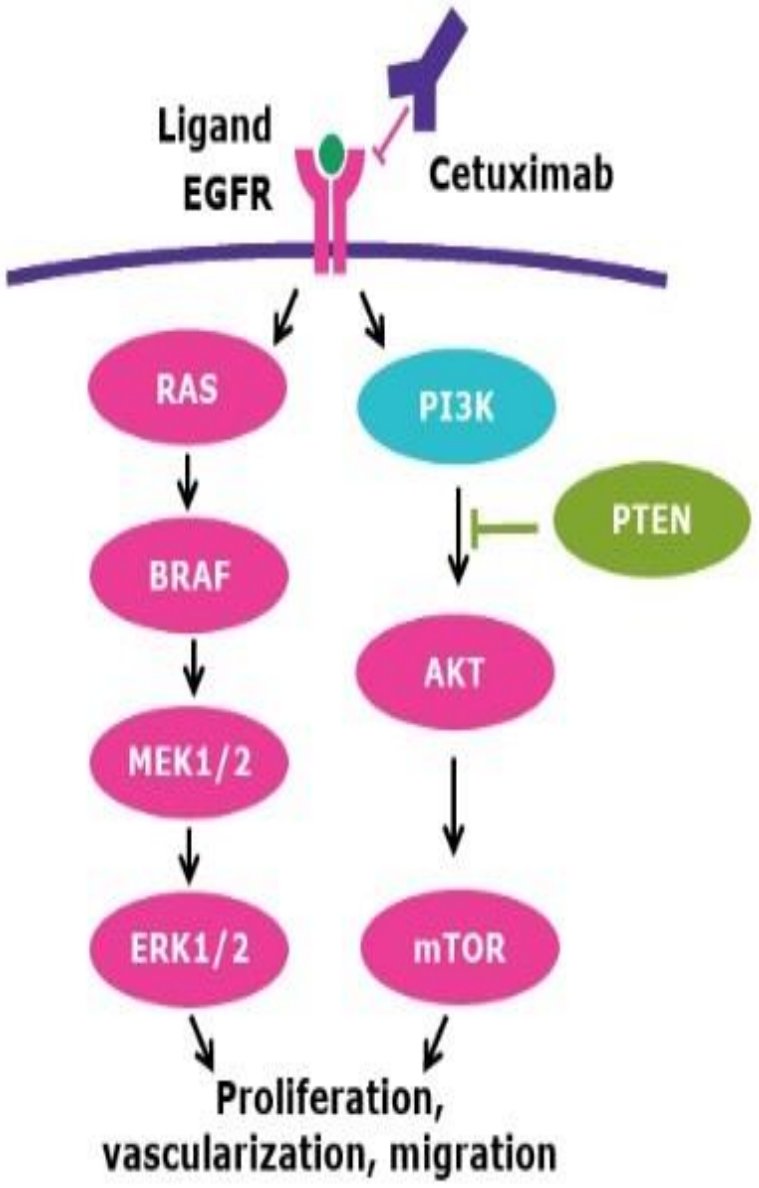


Biomarkers are gaining importance in the management of mCRC



PTEN loss: An alternative activator of PI3K

- PTEN antagonizes the action of PI3K[1]
- Loss of PTEN function increases PI3K/AKT signaling and is implicated in a variety of tumor types[1]
- Loss of PTEN function may occur via:[1,2]
  - Genomic alterations (point mutations/chromosomal deletions)
  - Epigenetic silencing (via promoter hypermethylation)
  - Transcriptional and post-transcriptional silencing
  - Post-translational regulation
  - Protein-protein interaction
- PTEN inactivation in CRC is mostly via promoter hypermethylation, combined with mutations or chromosomal deletion[1]



Unclear predictive impact of PTEN loss

Association between PTEN loss and/or mutation and poor outcomes in EGFR inhibitor-treated patients (KRAS [exon 2] wt)

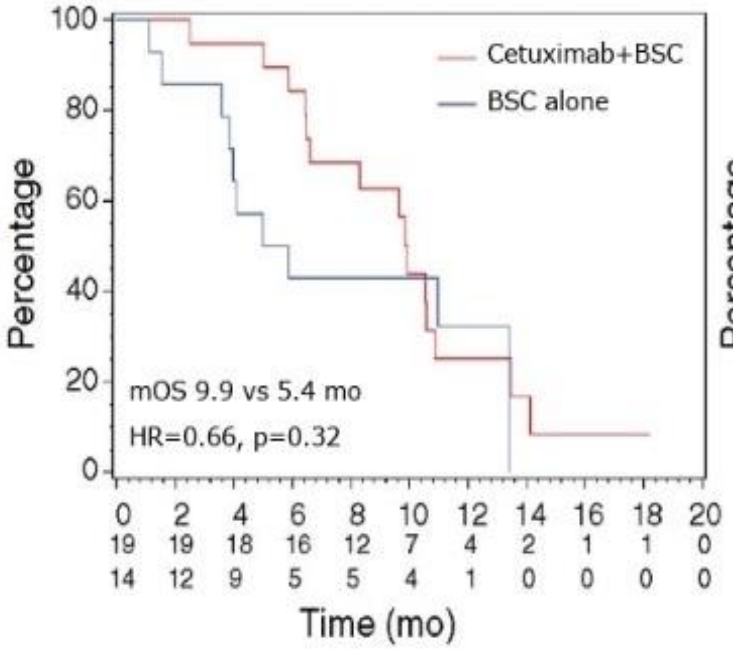
Meta-analysis	No. studies (patients, n)†	PTEN assessment	Outcome (PTEN-negative/mt vs PTEN-positive/wt)		
			ORR	PFS	OS
			RD (95% CI), p-value	HR (95% CI), p-value	HR (95% CI), p-value
Yang et al[1]	8 studies (n=591)	Protein expression	**-0.41 (-0.68–-0.14) p=0.003**	**1.75 (1.19–2.56) p=0.004**	**1.85 (1.30–2.64) p=0.0007**
			OR (95% CI)	HR (95% CI)	HR (95% CI)
Therkildsen et al[2*]	8 studies (n=NR)	Protein expression or gene mutation	**0.37 (0.18–0.73)**	**1.86 (1.37–2.52)**	**2.00 (1.49–2.69)**

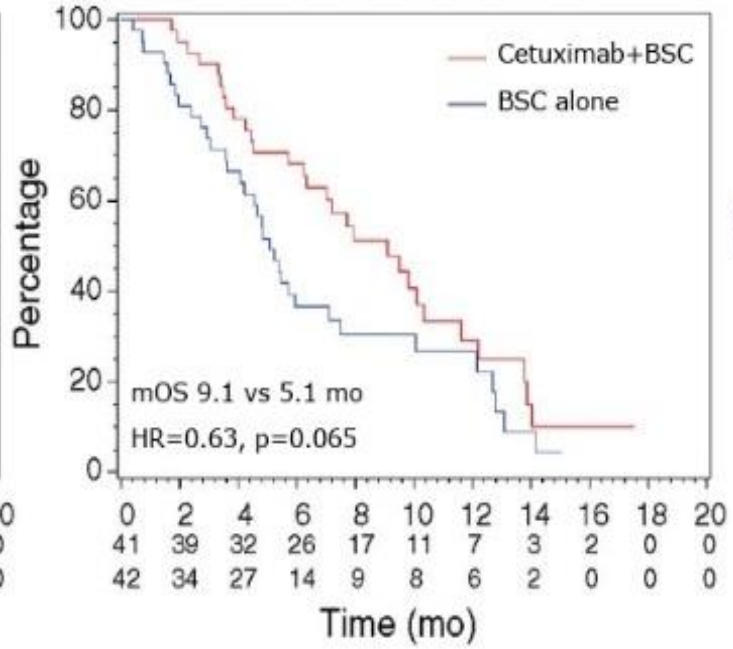
- Absence of comparator arms in these analyses precludes assessment of possible predictive relevance

Unclear predictive impact of PTEN loss

No significant OS or PFS benefit from cetuximab in tumors with PTEN loss in chemotherapy-refractory CRC (KRAS exon 2 wt) (NCIC CO.17)

OS in PTEN-positive





OS in PTEN-negative

Time (mo)

	Predictive analysis *in KRAS* WT subset adj HR (CET+BSC vs BSC), interaction p value	
	OS	PFS
BRAF	1.39, p=0.69	1.18, p=0.84
PI3KCA	0.79, p=0.63	0.73, p=0.50
PTEN	0.75, p=0.61	2.47, p=0.08

Signal Transduction and Targeted Therapy www.nature.com/sigtrans

REVIEW ARTICLE OPEN

Comprehensive review of targeted therapy for colorectal cancer

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Colorectal cancer (CRC) is among the most lethal and prevalent malignancies in the world and was responsible for nearly 881,000 cancer-related deaths in 2018. Surgery and chemotherapy have long been the first choices for cancer patients. However, the prognosis of CRC has never been satisfying, especially for patients with metastatic lesions. Targeted therapy is a new optional approach that has successfully prolonged overall survival for CRC patients. Following successes with the anti-EGFR (epidermal growth factor receptor) agent cetuximab and the anti-angiogenesis agent bevacizumab, new agents blocking different critical pathways as well as immune checkpoints are emerging at an unprecedented rate. Guidelines worldwide are currently updating the recommended targeted drugs on the basis of the increasing number of high-quality clinical trials. This review provides an overview of existing CRC-targeted agents and their underlying mechanisms, as well as a discussion of their limitations and future trends.

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Table 1. Agents targeting EGFR and EGFR-related pathways in colorectal cancer

Agent	Key trial (NCT number)	Design (*N*)	Subject	Treatment	Main results		
					RR	OS	PFS
EGFR inhibitor							
Cetuximab	Crystal[80]	Phase III	mCRC	FOLFIRI + CETU	46.9%	19.9 m	8.9 m
	NCT00154102	(*N* = 364)			(OR 1.4*)	(HR 0.93)	(HR 0.85*)
				FOLFIRI	38.7%	18.6 m	8.0 m
Cetuximab	Tailor[98]	Phase III	mCRC (RAS-WT)[a] First line	FOLFOX + CETU	61.1%	20.7 m	9.2 m
	NCT01228734	(*N* = 393)			(OR 2.41**)	(HR 0.76*)	(HR 0.69**)
				FOLFOX	39.5%	17.8 m	7.4 m
Panitumumab	PRIME[89,90]	Phase III	mCRC (KRAS-WT)	FOLFOX + PAN	57%	23.9 m	10.0 m
	NCT00364013	(*N* = 656)			(OR 1.47*)	(HR 0.88)	(HR 0.8*)
				FOLFOX	48%	19.7 m	8.6 m
Cetuximab	EPIC[94]	Phase III	mCRC	Cetuximab + Irinotecan	16.4%	10.7 m	4.0 m
		(*N* = 1298)	Second line			(HR 0.975)	(HR 0.692***)
				Irinotecan	4.2%	10 m	2.6 m
Panitumumab	PICCOLO[96]	Phase III	mCRC (KRAS-WT)	Panitumumab + Irinotecan	34%	10.4 m	NR
	ISRCTN	(*N* = 460)			(OR 4.12***)	(HR 1.01)	(HR 0.78*)
	93248876		Second line	Irinotecan	12%	10.5 m	NR
Panitumumab	20050181[508]	Phase III	mCRC (KRAS-WT)	FOLFIRI + Panitumumab	35%	14.5 m	5.9 m
		(*N* = 597)				(HR 0.85)	(HR 0.73**)
			Second line	FOLFIRI	10%	12.5 m	3.9 m
BRAF inhibitor with MEK inhibitor							
Vemurafenib	SWOG S1406[111]	Phase II	mCRC	Irinotecan + Cetuximab + Vemurafenib	16%	4.4 m	NR
	NCT02164916	(*N* = 106)	$BRAF^{V600}$ mutated				
			Extended RAS-WT			(HR 0.42***)	
			Treatment refractory	Irinotecan + Cetuximab	4%	2.0 m	NR
Dabrafenib and trametinib (MEK inhibitor)	Corcoran[109]	Phase I/II	mCRC	Dabrafenib + Trametinib	12%	NR	3.5 m
		(*N* = 43)	$BRAF^{V600}$ mutated				
Encorafenib + binimetinib (MEK inhibitor)	BEACON[113]	Phase III (safety lead-in results)	mCRC	ENCO + BINI + CETU	26%***	9 m	4.3 m
	NCT02928224		$BRAF^{V600E}$ mutated			(HR=0.52***)	(HR 0.40***)
			Second or third line	ENCO+CETU	20%***	8.4 m	4.2 m
		(*N* = 30)		Irinotecan/FOLFIRI + CETU		(HR=0.60***)	(HR=0.38***)
					2%	5.4 m	1.5 m
HER2 inhibitor							
Trastuzumab + pertuzumab	MyPathway[124]	Phase IIa	mCRC	Trastuzumab + pertuzumab	32%	11.5 m	2.9 m
	NCT02091141	(*N* = 57)	HER2 amplified				
Trastuzumab + lapatinib	HERACLES[125]	Phase II	mCRC	Trastuzumab+lapatinib	30%	46 weeks	21 weeks
		(*N* = 27)	HER2 positive				
			KRAS exon 2 WT				
			Treatment refractory				

Table 3. Antiangiogenic agents in colorectal cancer

Agent	Key trial (NCT number)	Design (*N*)	Subject	Treatment	Main results		
					RR	OS	PFS
Bevacizumab	CAIRO-3[186,197]	Phase III (*N* = 558)	mCRC	Capecitabine + Beva	NA	25.9 m	11.7 m
	NCT00442637		First-line treatment		NA	(HR 0.83)	(HR 0.67***)
				Observation		22.4 m	8. 5 m
Bevacizumab	TRIBE[187]	Phase III (*N* = 508)	mCRC	FOLFOXIRI + Beva	NA	29.8 m	12.3 m
	NCT00719797						
				FOLFIRI + Beva		(HR 0.8*)	(HR 0.77**)
					NA	25.8 m	9.7 m
Bevacizumab	AVEX[185]	Phase III (*N* = 280)	mCRC	Beva + capecitabine	19%	20.7 m	9.1 m
	NCT00484939			x		(HR 0.79)	(HR 0.53***)
				Capecitabine	10%	16.8 m	5.1 m
Bevacizumab	AVF2017[182]	Phase III (*N* = 813)	mCRC	Beva + IFL	44.8%	20.3 m	10.6 m
			Untreated			(HR 0.66**)	(HR 0.54**)
				Placebo + IFL	34.8%	15.6 m	6.2 m
Bevacizumab	ECOG3200[194]	Phase III (*N* = 829)	mCRC	FOLFOX + Beva	22.7%	12.9 m	7.3 m
			Second line			(HR 0.75**)	(HR 0.61***)
				FOLFOX	8.6%	10.8 m	4.7 m
				Beva	3.3%	10.2 m	2.7 m
Bevacizumab	ML18147[195]	Phase III (*N* = 810)	mCRC	Beva + Chemo[a]	2.8%	11.2 m	5.7 m
	NCT00700102		Second line	Chemo		(HR 0.81**)	(HR 0.68***)
					2.0%	9.8 m	4.1 m
Regorafenib	CONCUR[209]	Phase III (*N* = 204)	mCRC	Regorafenib	4%	8.8 m	3.2 m
	NCT01584830		Treatment refractory	Placebo		(HR 0.55***)	(HR 0.31***)
					0%	6.3 m	1.7 m
Regorafenib	CORRECT[208]	Phase III (*N* = 760)	mCRC	Regorafenib	1% (no CR)	6.4 m	1.9 m
	NCT01103323		Treatment refractory	Placebo		(HR 0.77**)	(HR 0.49***)
					0.4% (no CR)	5.0 m	1.7 m
Ziv-aflibercept	VELOUR[203]	Phase III (*N* = 1226)	mCRC	FOLFIRI + aflibercept	19.8%	13.5 m	6.9 m
	NCT00561470		Treatment refractory			(HR 0.817**)	(HR 0.76***)
				FOLFIRI + placebo	11.1%	12.06 m	4.67 m
Ramucirumab	RAISE[205]	Phase III (*N* = 1072)	mCRC	Ramucirumab + FOLFIRI	13.4%	13.3 m	5.7 m
	NCT01183780		Treatment refractory			(HR 0.844*)	(HR 0.793***)
				FOLFIRI + placebo			
					12.5%	11.7 m	4.5 m

Table 6. Main agents for immune checkpoint blockade therapy in colorectal cancer

Agent	Key trial (NCT number)	Design (*N*)	Subject	Main results		
				RR	OS (12 m)	PFS (12 m)
PD-1 plus CTLA-4 inhibitor						
Nivolumab + ipilimumab	CheckMate-142[382,398]	Phase II (*N* = 119)	dMMR/MSI-H mCRC	55%	85%	71%
	NCT02060188		Treatment refractory			
	CheckMate-142[382,399]	Phase II (*N* = 45)	dMMR/MSI-H mCRC	60%	83%	77%
	NCT02060188		Treatment refractory			
PD-1 inhibitor						
Nivolumab	CheckMate-142[382]	Phase II (*N* =74)	dMMR/MSI-H mCRC	31.1%	73.4%	50.4%
	NCT02060188		Treatment refractory			
Pembrolizumab	KEYNOTE-164[394,395]	Phase II (*N* = 61)	dMMR/MSI-H mCRC	33%	31.4 m	2.1 m
	NCT02460198		Treatment refractory			

mCRC metastatic colorectal cancer, *RR* response rate, *OS* overall survival, *PFS* progression-free survival, *dMMR* deficient mismatch repair, *MSI-H* microsatellite instability-high, *CTLA-4* cytotoxic T lymphocyte-associated antigen 4, *PD-1* programmed death-1, *m* months

Table 3. Antiangiogenic agents in colorectal cancer

Agent	Key trial (NCT number)	Design (*N*)	Subject	Treatment	Main results		
					RR	OS	PFS
Bevacizumab	CAIRO-3[186,197]	Phase III (*N* = 558)	mCRC	Capecitabine + Beva	NA	25.9 m	11.7 m
	NCT00442637		First-line treatment		NA	(HR 0.83)	(HR 0.67***)
				Observation		22.4 m	8. 5 m
Bevacizumab	TRIBE[187]	Phase III (*N* = 508)	mCRC	FOLFOXIRI + Beva	NA	29.8 m	12.3 m
	NCT00719797						
				FOLFIRI + Beva		(HR 0.8*)	(HR 0.77**)
					NA	25.8 m	9.7 m
Bevacizumab	AVEX[185]	Phase III (*N* = 280)	mCRC	Beva + capecitabine	19%	20.7 m	9.1 m
	NCT00484939			x		(HR 0.79)	(HR 0.53***)
				Capecitabine	10%	16.8 m	5.1 m
Bevacizumab	AVF2017[182]	Phase III (*N* = 813)	mCRC	Beva + IFL	44.8%	20.3 m	10.6 m
			Untreated			(HR 0.66**)	(HR 0.54**)
				Placebo + IFL	34.8%	15.6 m	6.2 m
Bevacizumab	ECOG3200[194]	Phase III (*N* = 829)	mCRC	FOLFOX + Beva	22.7%	12.9 m	7.3 m
			Second line			(HR 0.75**)	(HR 0.61***)
				FOLFOX	8.6%	10.8 m	4.7 m
				Beva	3.3%	10.2 m	2.7 m
Bevacizumab	ML18147[195]	Phase III (*N* = 810)	mCRC	Beva + Chemo[a]	2.8%	11.2 m	5.7 m
	NCT00700102		Second line	Chemo		(HR 0.81**)	(HR 0.68***)
					2.0%	9.8 m	4.1 m
Regorafenib	CONCUR[209]	Phase III (*N* = 204)	mCRC	Regorafenib	4%	8.8 m	3.2 m
	NCT01584830		Treatment refractory	Placebo		(HR 0.55***)	(HR 0.31***)
					0%	6.3 m	1.7 m
Regorafenib	CORRECT[208]	Phase III (*N* = 760)	mCRC	Regorafenib	1% (no CR)	6.4 m	1.9 m
	NCT01103323		Treatment refractory	Placebo		(HR 0.77**)	(HR 0.49***)
					0.4% (no CR)	5.0 m	1.7 m
Ziv-aflibercept	VELOUR[203]	Phase III (*N* = 1226)	mCRC	FOLFIRI + aflibercept	19.8%	13.5 m	6.9 m
	NCT00561470		Treatment refractory			(HR 0.817**)	(HR 0.76***)
				FOLFIRI + placebo	11.1%	12.06 m	4.67 m
Ramucirumab	RAISE[205]	Phase III (*N* = 1072)	mCRC	Ramucirumab + FOLFIRI	13.4%	13.3 m	5.7 m
	NCT01183780		Treatment refractory			(HR 0.844*)	(HR 0.793***)
				FOLFIRI + placebo			
					12.5%	11.7 m	4.5 m

CRC colorectal cancer, *mCRC* metastatic colorectal cancer, *RR* response rate, *OS* overall survival, *PFS* progression-free survival, *VEGF* vascular endothelial growth factor, *VEGFR* vascular endothelial growth factor receptor, *EGFR* epidermal growth factor receptor, *PDGFR* platelet-derived growth factor receptor, *FGFR* fibroblast growth factor receptor

Table 4. Antiangiogenic agents under clinical investigation

Name or ID	Targets	Condition	Phase	NCT identifier
LYN00101	VEGF	Solid tumors including CRC	Phase 1	NCT03644459
Vanucizumab	VEGF-A/angiopoietin-2	mCRC	Phase 2	NCT02141295
Sorafenib	VEGFR	mCRC	Phase 2	NCT03251612
		mCRC	Phase 2	NCT01471353
		Pretreated CRC	Phase 2	NCT01290926
		KRAS-mutated mCRC	Phase 2	NCT01715441
		mCRC	Phase 2	NCT00826540
		mCRC	Phase 2	NCT00865709
Linifanib	VEGFR	Advanced CRC	Phase 2	NCT00707889
Icrucumab	VEGFR	CRC	Phase 2	NCT01111604
Nintedanib	VEGFR	Refractory mCRC	Phase 3	NCT02149108
Vatalanib	VEGFR	mCRC	Phase 3	NCT00056446
		mCRC	Phase 3	NCT00056459
Semaxanib	VEGFR	mCRC	Phase 3	NCT00004252
		Advanced CRC	Phase 1/2	NCT00005818
Vandetanib	VEGFR/EGFR	CRC	Phase 2	NCT00454116
		mCRC	Phase 1	NCT00532090
		mCRC	Phase 2	NCT00500292
		Advanced CRC	Phase 1	NCT00496509
Famitinib	VEGFR-2/-3/PDGFR	Advanced CRC	Phase 2	NCT01762293
Tanibirumab	VEGFR-2	Advanced or metastatic cancer	Phase 1	NCT01660360
Cediranib	VEGFR-2	First-line mCRC	Phase 3	NCT00399035
		First-line mCRC	Phase 2/3	NCT00384176
		Solid tumors	Phase 2	NCT003851614
Brivanib	VEGFR-2/FGFR	KRAS-wild-type tumors and mCRC	Phase 3	NCT 00640471
LY3022856	VEGFR-3	Advanced solid tumors	Phase 1	NCT 01288989
Apatinib	VEGFR-2/c-Kit/ Src	Stage IIIB or IIIC CRC	Phase 3	NCT 03365765
		Second-line CRC	Phase 2	NCT 03271255
		Refractory CRC	Phase 2	NCT 03190616
Fruquintinib	Pan-VEGFR	Advanced CRC	Phase 4	NCT04005066
Tivozanib	Pan-VEGFR	First-line mCRC	Phase 2	NCT01478594
Motesanib AMG 706	Pan-VEGFR	mCRC	Phase 1	NCT 00101894
Sulfatinib	Pan-VEGFR	Advanced solid tumors	Phase 1	NCT 02133157
		Advanced solid tumors	Phase 1/2	NCT 02549937
Motesanib AMG 706	Pan-VEGFR	mCRC	Phase 1	NCT 00101894
Lenvatinib	Pan-VEGFR	Advanced solid tumors	Phase 2	NCT 03797326
Cabozantinib	Pan-VEGFR	Solid tumors	Phase 1/2	NCT03170960
		KRAS-wild-type mCRC	Phase 1	NCT02008383
Axitinib	Pan-VEGFR	First-line mCRC	Phase 2	NCT 01490866
		mCRC	Phase 2	NCT 00615056
Pazopanib	Pan-VEGFR	Second-line mCRC	Phase 1	NCT 00540943
		CRC	Phase 1	NCT00387387
Sunitinib	Pan-VEGFR	mCRC	Phase 3	NCT 00457691
MNRP1685A	Neuropilin-1	Solid tumors	Phase 1	NCT00747734
		Solid tumors	Phase 1	NCT00954642

CRC colorectal cancer, *mCRC* metastatic colorectal cancer, *VEGF* vascular endothelial growth factor, *VEGFR* vascular endothelial growth factor receptor, *EGFR* epidermal growth factor receptor, *PDGFR* platelet-derived growth factor receptor, *FGFR* fibroblast growth factor receptor

Table 5. HGF-MET-targeted agents under clinical investigation

Name or ID	Targets	Condition	Phase	NCT identifier
Rilotumumab	HGF	Gastrointestinal cancer	Phase 1/2	NCT00788957
TAK-701	HGF	Advanced solid tumors	Phase 1	NCT00831896
Onartuzumab	MET	CRC	Phase 2	NCT01418222
ABT-700	MET	Advanced solid tumors	Phase 1	NCT01472016
ABBV-399	MET	Solid tumors	Phase 1	NCT02055066
YYB-101	MET	Solid tumors	Phase 1	NCT02499224
ARGX-111	MET	Solid tumors	Phase 1	NCT02099058
Tivantinib	MET	mCRC	Phase 1/2	NCT01075048
Savolitinib	MET	mCRC	Phase 2	NCT03592641
AMG 337	MET	Advanced solid tumors	Phase 1	NCT01253707
Capmatinib	MET	Multiple tumors including CRC	Phase 1	NCT02386826
Crizotinib	MET/RON/ROS	Solid tumor	Phase 1	NCT02510001
		Solid neoplasm	Phase 2	NCT02465060
Cabozantinib	MET/RET/VEGFR-2	CRC	Phase 1	NCT02008383
		CRC	Phase 1	NCT03539822
		CRC	Phase 1	NCT03798626
		Multiple tumors including CRC	Phase 1/2	NCT03170960
		CRC	Phase 2	NCT03542877
Foretinib	MET/VEGFR	Solid tumors	Phase 1	NCT00742261
		Solid tumors	Phase 1	NCT00743067
Golvatinib	MET/VEGFR	Solid tumors	Phase 1/2	NCT01355302
Merestinib	MET/TEK/ROS1/DDR/MKNK	Multiple tumors including CRC	Phase 1	NCT02745769
Sitravatinib	MET/VEGFR/DDR	Advanced tumors	Phase 1	NCT02219711
		Advanced tumors	Phase 1	NCT03666143

CRC colorectal cancer, *mCRC* metastatic colorectal cancer, *HGF* hepatocyte growth factor, *MET* mesenchymal–epithelial transition factor, *TEK* tunica interna endothelial cell kinase, *DDR* discoidin domain receptor tyrosine kinase, *MKNK* MAP kinase-interacting serine/threonine protein kinase, *FLT3* Fms-related tyrosine kinase 3, *VEGFR* vascular endothelial growth factor receptor, *MEK* mitogen-activated protein kinase

Table 6. Main agents for immune checkpoint blockade therapy in colorectal cancer

Agent	Key trial (NCT number)	Design (*N*)	Subject	Main results		
				RR	OS (12 m)	PFS (12 m)
PD-1 plus CTLA-4 inhibitor						
Nivolumab + ipilimumab	CheckMate-142[382,398]	Phase II (*N* = 119)	dMMR/MSI-H mCRC	55%	85%	71%
	NCT02060188		Treatment refractory			
	CheckMate-142[382,399]	Phase II (*N* = 45)	dMMR/MSI-H mCRC	60%	83%	77%
	NCT02060188		Treatment refractory			
PD-1 inhibitor						
Nivolumab	CheckMate-142[382]	Phase II (*N* = 74)	dMMR/MSI-H mCRC	31.1%	73.4%	50.4%
	NCT02060188		Treatment refractory			
Pembrolizumab	KEYNOTE-164[394,395]	Phase II (*N* = 61)	dMMR/MSI-H mCRC	33%	31.4 m	2.1 m
	NCT02460198		Treatment refractory			

mCRC metastatic colorectal cancer, *RR* response rate, *OS* overall survival, *PFS* progression-free survival, *dMMR* deficient mismatch repair, *MSI-H* microsatellite instability-high, *CTLA-4* cytotoxic T lymphocyte-associated antigen 4, *PD-1* programmed death-1, *m* months

Table 7. Immune checkpoint modulators under clinical investigation

Name or ID	Targets	Condition	Phase	NCT identifier
Atezolizumab	PD-1/PD-L1	mCRC	Phase 3	NCT02788279
		Stage 3 CRC	Phase 3	NCT02912559
		First-line mCRC	Phase 3	NCT02997228
		Refractory CRC	Phase 2	NCT02873195
		First-line mCRC	Phase 2	NCT02291289
		mCRC	Phase 1	NCT02876224
Avelumab	PD-1/PD-L1	mCRC	Phase 2	NCT03150706
		mCRC	Phase 2	NCT03258398
PDR-001	PD-1/PD-L1	First-line mCRC	Phase 1	NCT03176264
		mCRC	Phase 1	NCT03081494
SHR-1210	PD-1/PD-L1	CRC/HCC/NSCLC	Phase 1	NCT03601598
JS-001 Toripalimab	PD-1/PD-L1	CRC	Phase 1/2	NCT03946917
		CRC	Phase 2	NCT04118933
		mCRC	Phase 2	NCT03927898
AMP-224	PD-1/PD-L1	CRC	Phase 1	NCT02298946
TSR-033	PD-1/PD-L1	CRC	Phase 1	NCT03250832
Camrelizumab	PD-1/PD-L1	mCRC	Phase 2	NCT03912857
AB-122	PD-1/PD-L1	Multiple tumors including CRC	Phase 1	NCT03629756
		Multiple tumors including CRC	Phase 1	NCT03628677
INT230-6	PD-1/CTLA-4	Multiple tumors including CRC	Phase 1/2	NCT03058289
ONC-392	CTLA-4	Multiple tumors including CRC	Phase 1/2	NCT04140526
Tremelimumab	CTLA-4	mCRC	Phase 1/2	NCT03202758
		CRC	Phase 1/2	NCT03206073
		mCRC	Phase 2	NCT03122509
		mCRC	Phase 2	NCT03428126
		mCRC	Phase 2	NCT02811497
		mCRC	Phase 2	NCT03122509
		mCRC	Phase 2	NCT03435107
		mCRC	Phase 1/2	NCT03202758
MGD-013	PD-1/LAG-3	Solid tumors	Phase 1	NCT03219268
Relatlimab	LAG-3	Solid tumors	Phase 1/2	NCT01968109
		CRC	Phase 2	NCT03642067
TSR-033	LAG-3	Solid tumors	Phase 1	NCT03250832
IMP-321	LAG-3	Solid tumors	Phase 1	NCT03252936
		Solid tumors	Phase 1	NCT02676869
REGN-3767	LAG-3	Solid tumors	Phase 1	NCT03005782
TSR-022	TIM-3	Solid tumors	Phase 1	NCT02817633
MBG-453	TIM-3	Advanced malignancies	Phase 1	NCT02608268
AB-154	TIGIT	Multiple tumors including CRC	Phase 1	NCT03628677
KRN-330	A33 glycoprotein	A33-positive colorectal cancer	Phase 1	NCT00575562
I-huA33	A33 glycoprotein	A33-positive colorectal cancer	Phase 1	NCT00291486

CRC colorectal cancer, *mCRC* metastatic colorectal cancer, *CTLA-4* cytotoxic T lymphocyte-associated antigen 4, *PD-1/PD-L1* programmed death-1/programmed death ligand 1, *TIM-3* T cell immunoglobulin and mucin domain-containing protein 3, *TIGIT* T cell immunoglobulin and ITIM domains, *LAG-3* lymphocyte activation gene 3

Table 8. Novel agents under clinical investigation

Name or ID	Targets	Condition	Phase	NCT identifier
WNT-974	Wnt	mCRC	Phase 1/2	NCT02278133
FOXY-5	Wnt	Multiple tumors including CRC	Phase 1	NCT02655952
		Multiple tumors including CRC	Phase 1	NCT02020291
		CRC	Phase 2	NCT03883802
LGK-974	Wnt	Multiple tumors including CRC	Phase 1	NCT01351103
RO4929097	Notch	CRC	Phase 1	NCT01198535
		CRC	Phase 2	NCT01116687
LY3039478	Notch	Solid tumors including CRC	Phase 1	NCT02784795
CB-103	Notch	Solid tumors including CRC	Phase 1/2	NCT03422679
MK-0752	Notch	Malignant neoplasms	Phase 1	NCT01243762
GEN-1029	DR5	Solid tumors including CRC	Phase 1/2	NCT03576131
INBRX-109	DR5	Solid tumors including CRC	Phase 1	NCT03715933
DS-8273a	DR5	CRC	Phase 1	NCT02991196
Conatumumab	DR5	mCRC	Phase 2	NCT00813605
		Solid tumors including CRC	Phase 2	NCT01327612
Vismodegib	Hedgehog	mCRC	Phase 2	NCT00636610
		Multiple tumors including CRC	Phase 2	NCT00959647
		First-line mCRC	Phase 1/2	NCT00625651
LDE225	Hedgehog	Solid tumors	Phase 1	NCT01576666
LY3200882	TGF	mCRC	Phase 1/2	NCT04031872
NIS-793	TGF	Multiple tumors including CRC	Phase 1	NCT02947165
KW-2450	IGF-1R	Advanced solid tumor	Phase 1	NCT00921336
IMC-A12	IGF-1R	mCRC	Phase 2	NCT00503685
Dalotuzumab	IGF-1R	mCRC	Phase 2	NCT00614393
		CRC	Phase 1	NCT00925015
Figitumumab	IGF-1R	Stage IV CRC	Phase 2	NCT00560560
Robatumumab	IGF-1R	Relapsed or recurrent CRC	Phase 2	NCT00551213
ganitumab	IGF-1R/HGF	KRAS-wild-type mCRC	Phase 1/2	NCT00788957
		mCRC	Phase 2	NCT00891930
		KRAS-mutant mCRC	Phase 2	NCT00813605

CRC colorectal cancer, *mCRC* metastatic colorectal cancer, *DR5* human death receptor 5, *TGF* transforming growth factor, *IGF-1R* insulin-like growth factor 1 receptor, *HGF* hepatocyte growth factor

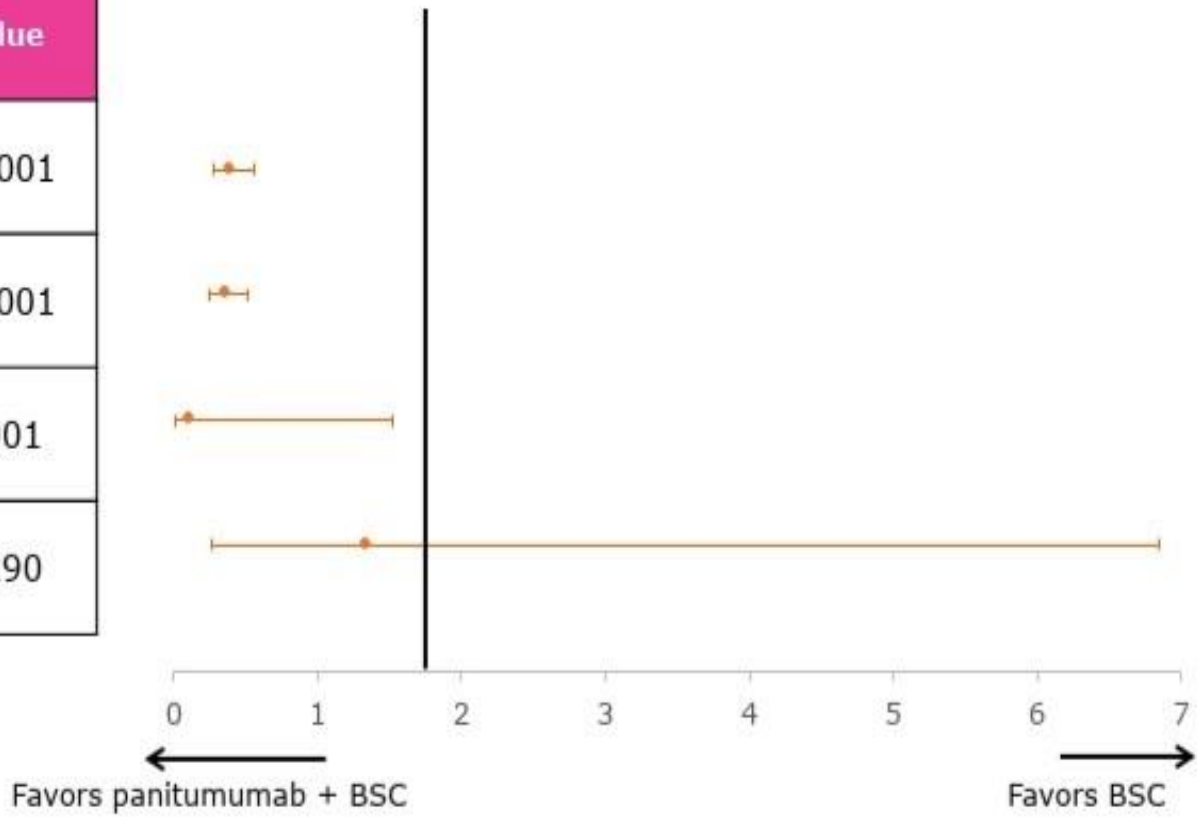
Which biomarkers are important in mCRC today?

Molecular biomarker	Prognostic [Evidence available][1]	Predictive [Evidence available] [1]
RAS mt	Prognostic for poor outcome [Insufficient]	Negative predictor of anti-EGFR response **[Conclusive]**
BRAF mt	Prognostic for poor outcome **[Substantial]**	Potential predictive marker for resistance [Insufficient]
PTEN	Not prognostic for outcome	Potential predictive marker for resistance [Insufficient]
HER2	Not prognostic for outcome	Potential predictive marker for resistance [Insufficient]

Unclear predictive impact of PTEN loss

PFS benefit of panitumumab vs BSC in chemorefractory KRAS wt mCRC appeared consistent irrespective of PTEN mutational status

PTEN status† (KRAS wt*)	n	Panitumumab + BSC vs BSC PFS HR (95% CI)	p-value
Unselected	153	0.39 (0.28–0.56)	<0.0001
wt	135	0.36 (0.25–0.52)	<0.0001
mt	9	0.11 (0.01–1.52)	0.1001
Not determined	9	1.34 (0.26–6.85)	0.7290



- Response rates with panitumumab + BSC in chemorefractory KRAS wt* mCRC were similar irrespective of PTEN mutational status

*KRAS exon 2 (codons 12 and 13) and exon 3 (codon 61)
†All PTEN exons were analyzed
Peeters M, et al. Clin Cancer Res 2013;19:1902–1912.

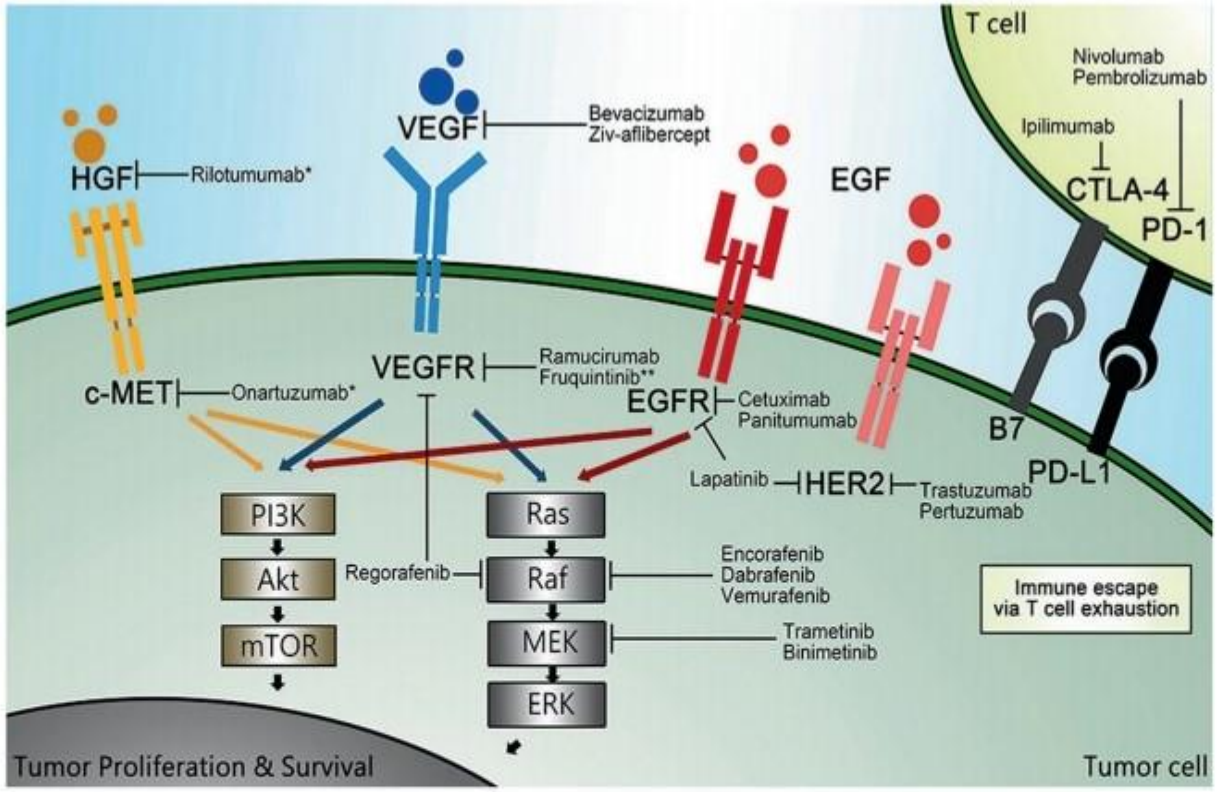


Fig. 3 Overview of National Comprehensive Cancer Network (NCCN)-recommended targeted agents. HGF: hepatocyte growth factor; c-MET: mesenchymal–epithelial transition factor; VEGF: vascular endothelial growth factor; VEGFR: vascular endothelial growth factor receptor; EGFR: epidermal growth factor receptor; EGF: epidermal growth factor; HER2: human epidermal growth factor 2; CTLA-4: cytotoxic T lymphocyte-associated antigen 4; PD-1: programmed death-1; PD-L1: programmed death ligand 1; PI3K: phosphoinositide 3-kinase; AKT: protein kinase B, also known as PKB; mTOR: mammalian target of rapamycin; MEK: mitogen-activated protein kinase; ERK: extracellular signal-regulated kinase. *These agents have not been recommended by the NCCN. **This agent has been approved by the National Medical Products Administration of China (NMPA), but not by the United States of America Food and Drug Administration (FDA)

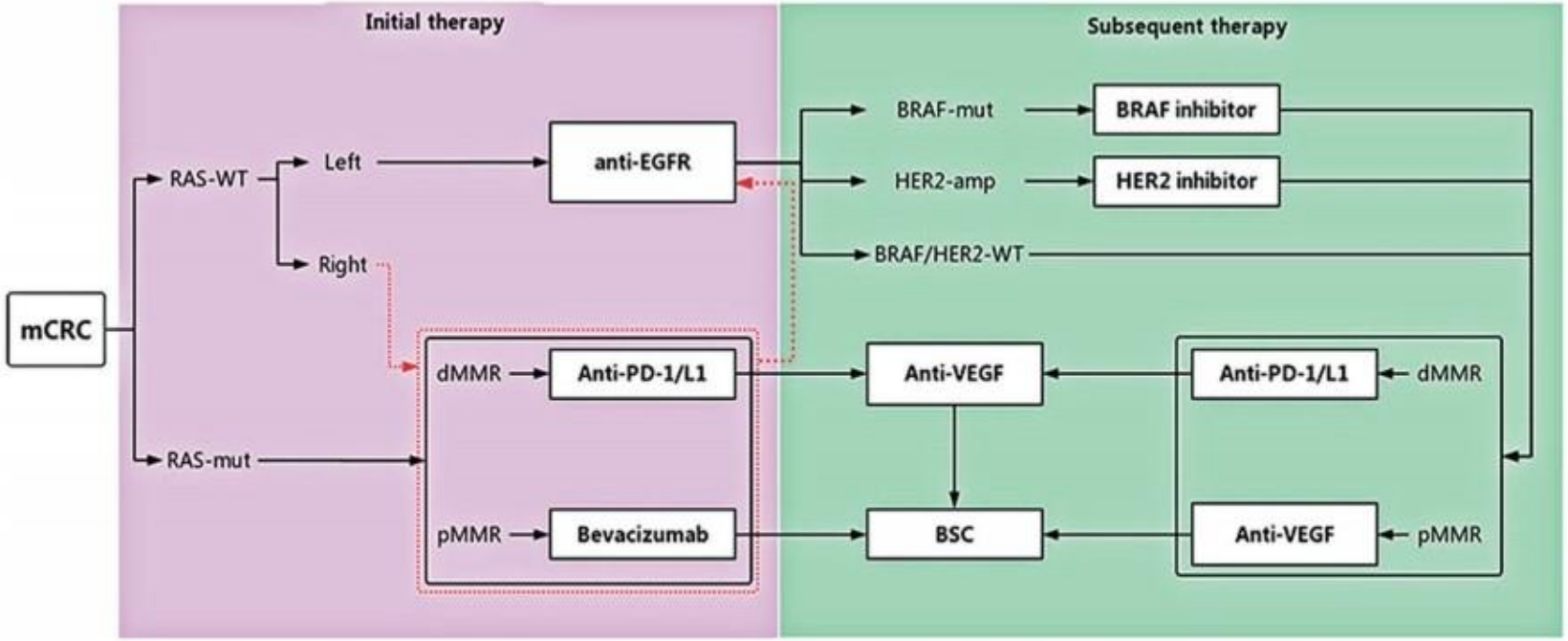


Fig. 4 National Comprehensive Cancer Network (NCCN)-recommended strategy for metastatic colorectal cancer targeted therapy. mCRC: metastatic colorectal cancer; EGFR: epidermal growth factor receptor; VEGF: vascular endothelial growth factor; PD-1/L1: programmed death-1/programmed death ligand 1; dMMR: deficient mismatch repair; pMMR: proficient mismatch repair; HER2: human epidermal growth factor 2; BSC: best supportive care; WT: wild type; mut: mutated; amp: amplified. *The NCCN recommends initial administration of PD-1/PD-L1 therapy only in patients in poor functional status

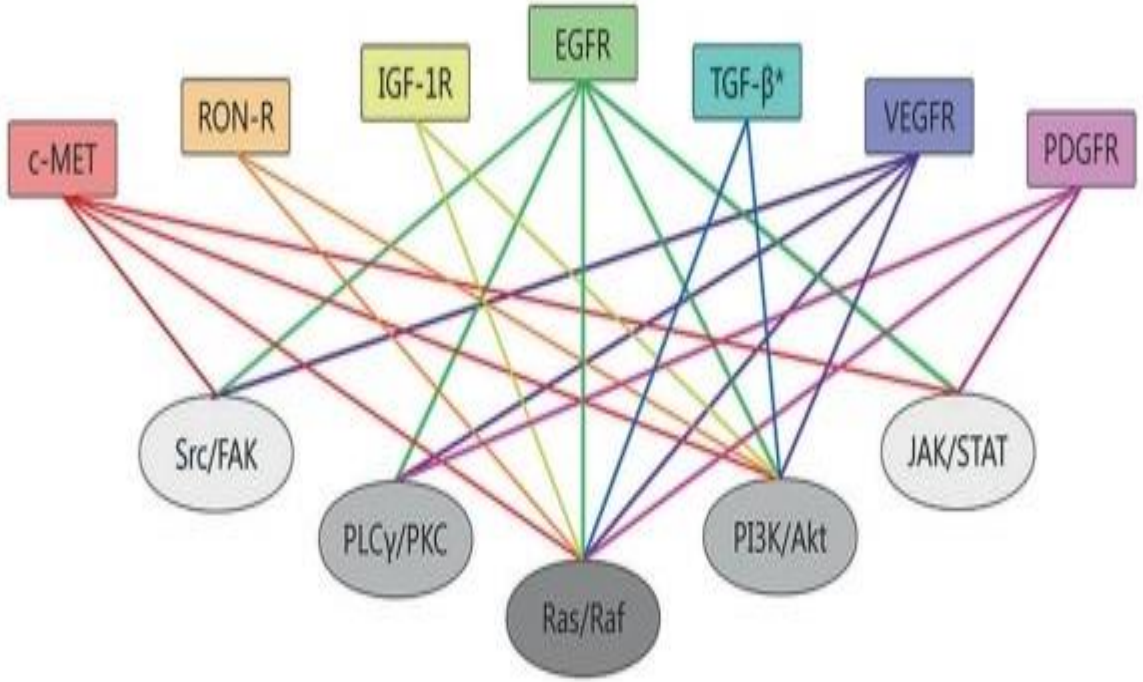
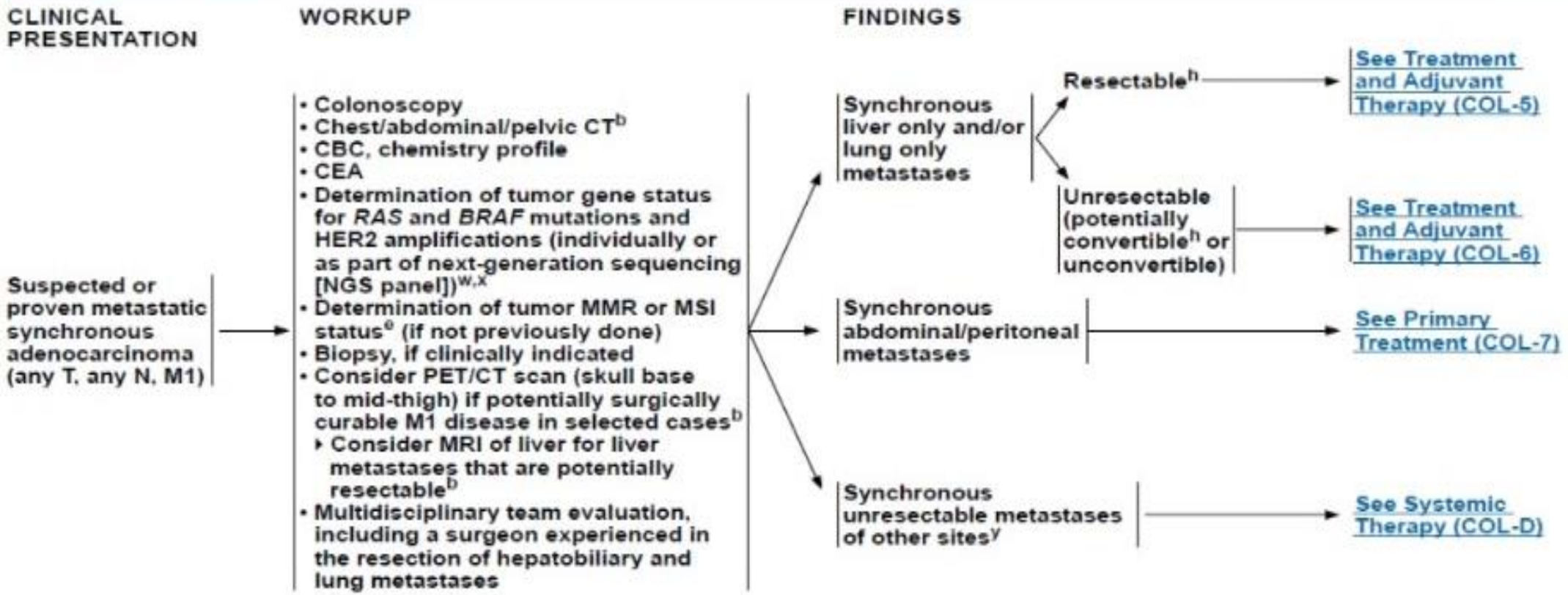


Fig. 5 Crosstalk and bypass mechanisms between pathways. VEGFR: vascular endothelial growth factor receptor; EGFR: epidermal growth factor receptor; c-MET: mesenchymal–epithelial transition factor; IGF-1R: insulin-like growth factor 1 receptor; TGF-β: transforming growth factor-β; RON-R: recepteur d'Origine nantais; PDGFR: platelet-derived growth factor receptor; *with multiple isoforms



[b] See Principles of Imaging (COL-A).
[e] See Principles of Pathologic Review (COL-B 4 of 8) - MSI or MMR Testing.
[h] See Principles of Surgery (COL-C 2 of 3).
[w] See Principles of Pathologic Review (COL-B 4 of 8) - *KRAS*, *NRAS*, and *BRAF* Mutation Testing.
[x] If known *RAS/RAF* mutation, HER2 testing is not indicated. NGS panels have the ability to pick up rare and actionable mutations and fusions.
[y] Consider colon resection only if imminent risk of obstruction, significant bleeding, perforation, or other significant tumor-related symptoms.

Note: All recommendations are category 2A unless otherwise indicated.
Clinical Trials: NCCN believes that the best management of any patient with cancer is in a clinical trial. Participation in clinical trials is especially encouraged.

TREATMENT
Unresectable[h] synchronous liver and/or lung metastases only

- Systemic therapy
  - FOLFIRI or FOLFOX or CAPEOX or FOLFOXIRI ± bevacizumab[bb,cc]
    or
  - FOLFIRI or FOLFOX or FOLFOXIRI ± panitumumab or cetuximab[dd] (category 2B for FOLFOXIRI combination) (*KRAS/NRAS/BRAF* WT gene and left-sided tumors only)[w,ee]
- Consider colon resection[h] only if imminent risk of obstruction, significant bleeding, perforation, or other significant tumor-related symptoms

→ Re-evaluate for conversion to resectable[b,h] every 2 mo if conversion to resectability is a reasonable goal

→ Converted to resectable → Synchronized or staged resection[h] of colon and metastatic cancer

ADJUVANT TREATMENT[b]

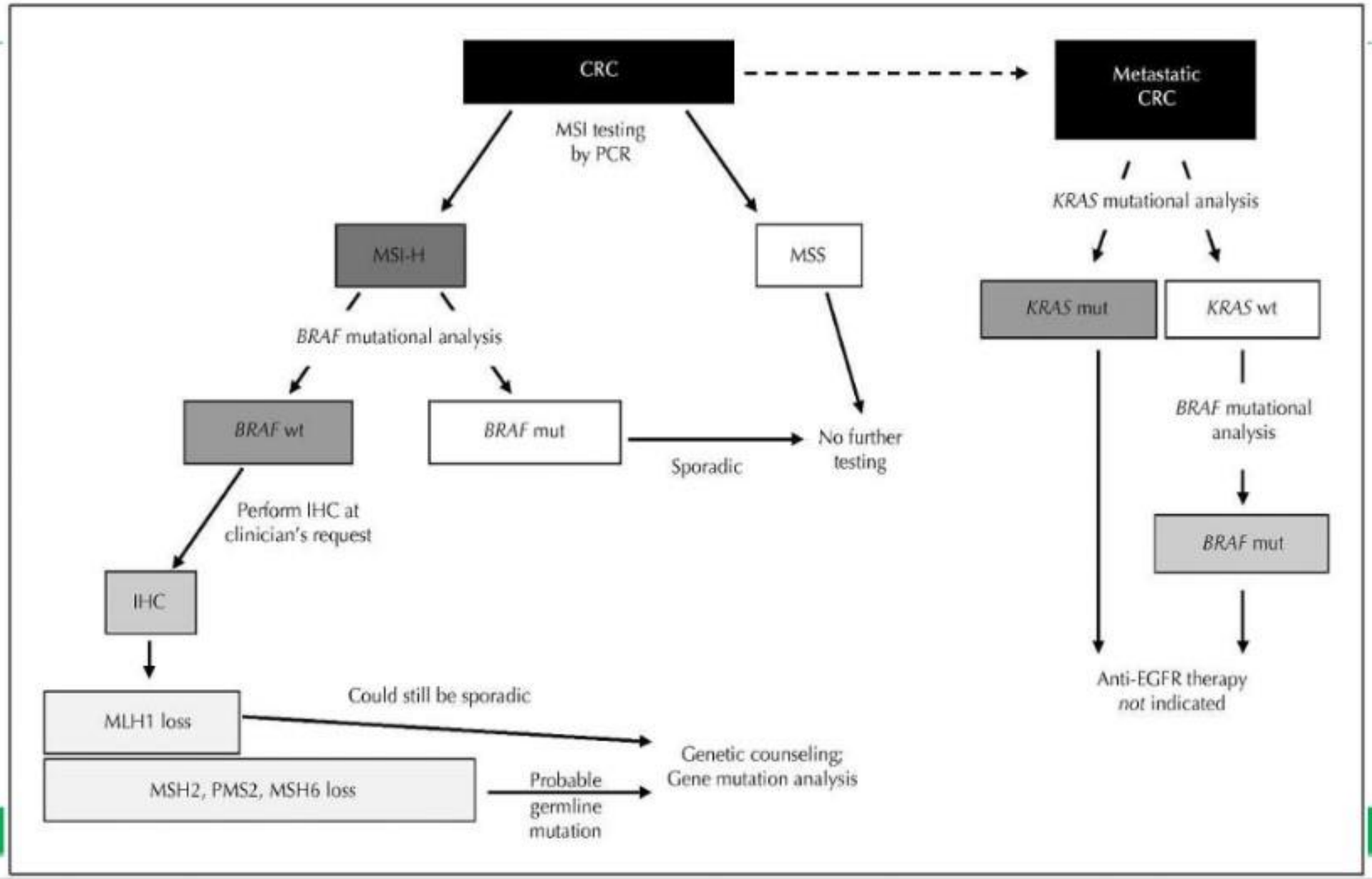
→ Systemic therapy ± biologic therapy[ff] (COL-D) (category 2B for biologic therapy)
or
Consider observation or shortened course of chemotherapy (6 MO TOTAL PERIOPERATIVE TREATMENT PREFERRED)

→ See Surveillance (COL-8)

→ Remains unresectable → See Systemic Therapy (COL-D)

See Recurrence (COL-9)

[b] See Principles of Imaging (COL-A).
[h] See Principles of Surgery (COL-C 2 of 3).
[w] See Principles of Pathologic Review (COL-B 4 of 8) - *KRAS*, *NRAS*, and *BRAF* Mutation Testing.
[bb] There should be at least a 6-week interval between the last dose of bevacizumab and elective surgery and re-initiation of bevacizumab at least 6–8 weeks postoperatively. There is an increased risk of stroke and other arterial events, especially in those aged ≥65 years. The use of bevacizumab may interfere with wound healing.
[cc] An FDA-approved biosimilar is an appropriate substitution for bevacizumab.
[dd] There are conflicting data regarding the use of FOLFOX + cetuximab in patients who have potentially resectable liver metastases.
[ee] The panel defines the left side of the colon as splenic flexure to rectum. Evidence suggests that patients with tumors originating on the right side of the colon (hepatic flexure through cecum) are unlikely to respond to cetuximab and panitumumab in first-line therapy for metastatic disease. Data on the response to cetuximab and panitumumab in patients with primary tumors originating in the transverse colon (hepatic flexure to splenic flexure) are lacking.
[ff] Biologic therapy is only appropriate for continuation of favorable response from conversion therapy.

Note: All recommendations are category 2A unless otherwise indicated.
Clinical Trials: NCCN believes that the best management of any patient with cancer is in a clinical trial. Participation in clinical trials is especially encouraged.

Personalized Cancer Medicine
/Targeted therapy/