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Tumour Review

Diagnosis and treatment of human epidermal growth factor receptor 2-positive metastatic colorectal cancer: a systematic literature review

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ABSTRACT

Human epidermal growth factor receptor 2 (HER2)-targeted therapies have been investigated for therapeutic benefit in *RAS/BRAF* wild-type/HER2+ metastatic colorectal cancer (mCRC). Unlike HER2+ breast and gastric cancer, there are no regulatory criteria for determining HER2 overexpression in patients with mCRC. This systematic literature review describes unmet needs for patients with HER2+ mCRC in relation to testing and treatment, highlights the importance of early HER2 testing at mCRC diagnosis, and discusses the evolving treatment landscape. We utilised PubMed and EMBASE databases up to November 2023 to identify journal articles and published congress abstracts relating to the HER2+ disease and treatment landscape, HER2 testing in mCRC, and HER2-targeted treatments in mCRC. Many studies have demonstrated the utility of immunohistochemistry, *in situ* hybridisation, and next-generation sequencing (tissue- and circulating tumour DNA-based) for detecting HER2 overexpression/amplification in mCRC and have attempted to establish consolidated criteria like those used for breast and gastric cancer. The value of HER2-targeted treatments in patients with HER2+ mCRC has been evidenced by clinical trials and meta-analyses, with strong evidence from MOUNTAINEER and DESTINY-CRC01 which supports the use of tucatinib + trastuzumab or trastuzumab deruxtecan, respectively, among this patient population. However, real-world analyses have confirmed that patients with HER2+ mCRC are not routinely tested for HER2 overexpression. Strong evidence and clinical guidelines support the value of HER2-targeted treatment among patients with HER2+ mCRC. There is a need for increased awareness and earlier uptake of HER2 testing among patients with mCRC to broaden treatment options and optimise outcomes for this patient population.

Introduction

Globally, colorectal cancer (CRC) is the third most common cancer and the second most common cause of cancer-related death [1]. In 2022, there were over 1.9 million new cases of CRC and it was associated with 903,859 deaths [1]. Approximately 15%–30% of patients have metastatic disease at diagnosis of CRC. Of those diagnosed with localised disease, up to 50% will eventually develop metastases [2]. The prognosis of patients with metastatic CRC (mCRC) has vastly improved over the last 20 years [3], although the increasing incidence of mCRC among individuals younger than 50 years of age represents a new challenge of preclinical and clinical research [4]. The identification of clinically

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relevant molecular factors (e.g. mismatch repair status; *KRAS*, *NRAS*, and *BRAF* mutations; and rare translocations of *NTRK1/2/3* genes) has been shifting the mCRC treatment landscape away from conventional chemotherapy-based regimens towards targeted therapies and immunotherapies [2].

Human epidermal growth factor receptor 2 (HER2) was first investigated as a potential therapeutic target in breast and gastric cancer during the 1980s [5–7]. HER2 overexpression occurs in 15%–30% and 10%–30% of patients with advanced breast and gastric cancer, respectively [7–9]. The establishment of HER2 as an actionable target in breast and gastric cancer pioneered the development of HER2-targeted treatments, and trastuzumab was the first US Food and Drug Administration (FDA)- and European Medicines Agency (EMA)-approved treatment for HER2+ breast and gastric cancer [10,11]. HER2-targeted therapies have significantly improved clinical outcomes for patients with HER2+ breast and gastric cancer, and the European Society for Medical Oncology (ESMO) guidelines and NCCN Clinical Practice Guidelines in Oncology (NCCN Guidelines®) include HER2 testing in diagnostic recommendations for both tumour types [12–16]. Owing to differences in the biology of HER2 expression in breast and gastric cancers (e.g. immunohistochemistry [IHC] staining distribution, intratumoural heterogeneity, impact of differentiation, and anatomic location), there are distinct HER2 testing criteria for each tumour type [17–21].

More recently, HER2 expression has been investigated in patients with mCRC. The use of HER2 as an actionable target in mCRC was initially explored in phase II trials of trastuzumab combined with chemotherapy [22–24]. However, evaluations in mCRC were delayed due to the lack of preclinical models indicating an optimal pharmacological strategy and the infrequent occurrence of *HER2* alterations among patients with mCRC [22]. Consequently, optimal treatment strategies and testing methods are less established in mCRC than in other cancers.

The aims of this systematic literature review are to describe the unmet needs for patients with HER2+ mCRC in relation to testing and treatment options, raise awareness of the importance of early HER2 testing at diagnosis of mCRC, and discuss the evolving treatment landscape in HER2+ mCRC.

Methods

Literature search strategy

We performed a systematic literature search in line with Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines [25] to identify relevant information on unmet needs, HER2 testing methodologies, and treatment options for patients with HER2+ mCRC. We performed three targeted searches of the PubMed database (https://pubmed.ncbi.nlm.nih.gov/) for relevant journal articles and a search of the EMBASE database (https://www.embase.com/landing?status=grey) for relevant abstracts from key congresses.

PubMed search

On 1 November 2023, we conducted a 20-year general search of the HER2+ disease state and treatment landscape, a 10-year focused search on HER2 testing in mCRC, and a 10-year focused search on HER2-targeted treatments in mCRC. All search strings are outlined in **Supplementary Table S1**.

EMBASE congress search

We searched the EMBASE database for abstracts from the past 5 years across six key congresses: American Association for Cancer Research, American Society of Clinical Oncology (ASCO), ASCO Gastrointestinal Cancers Symposium, ESMO, ESMO Gastrointestinal Cancers Congress, and United States and Canadian Academy of Pathology. The same search string was used for all congresses (outlined in **Supplementary Table S1**).

Filtering process

All results from the three PubMed searches and the EMBASE congress search were extracted into an Excel spreadsheet for filtering. Duplicate entries were removed using the automated Excel function.

To identify publications for full-text analysis, all results were manually reviewed for relevance against predefined eligibility criteria in a series of triaging steps (**Supplementary Table S2**). In screening pass 1, the titles of all PubMed articles were screened for relevance and articles that did not meet the inclusion criteria were excluded. In screening pass 2, the abstracts of the included PubMed articles and all EMBASE congress hits were screened for relevance; any articles that did not meet the inclusion criteria were excluded. To avoid selection bias, a second reviewer screened the selections.

Data extraction

All publications that passed the screening phases were analysed and the following information was extracted: key publication information (including publication type and audience); information relating to the biology of HER2+ mCRC, HER2 testing in HER2+ mCRC (including discussed testing methods), or the treatment landscape for HER2 + mCRC (including assessed patient population, eligibility criteria, efficacy outcomes, safety outcomes, and drug mechanism of action); and any other comments relevant to the discussion.

Any publications that did not contain any relevant information for extraction were excluded. In cases where there were multiple publications that reported the same analysis for the same trial, the most recent congress presentation or full publication was prioritised, and others were excluded. Alongside the outputs of the literature search, this report includes some additional publications based on author awareness of key analyses and clinical trials within HER2+ mCRC to supplement the discussion. To bolster the findings of the literature search, we additionally provide some author guidance regarding an optimal testing and treatment strategy for HER2+ mCRC.

Results

Literature search overview

Fig. 1 outlines our literature search filtering process. Across the three targeted PubMed searches and the EMBASE congress abstract search, there were a total of 352 initial hits. After removal of duplicate entries, we screened 96 PubMed hits and 145 EMBASE congress hits, and performed full-text analysis of 59 publications from the literature search. In total, 71 journal articles or congress abstracts were identified for inclusion in the final report.

Overview of HER2 alterations in mCRC

HER2 overexpression in mCRC

Table 1 presents key findings from our literature search relating to the frequency, associated clinical characteristics, and prognostic impact of HER2 overexpression in mCRC. In summary, HER2 overexpression is observed in a relatively small subset of patients with CRC (4%), with a higher frequency in the distal colon and rectum (left-sided tumours), and among patients with *KRAS* or *RAS/BRAF* wild-type (wt) disease [26–31]. The prognostic impact of HER2 overexpression in mCRC has not yet been fully elucidated [32,33].

HER2 mutations in mCRC

Table 1 presents key findings from our literature search relating to the occurrence of *HER2* mutations in mCRC, and their association with

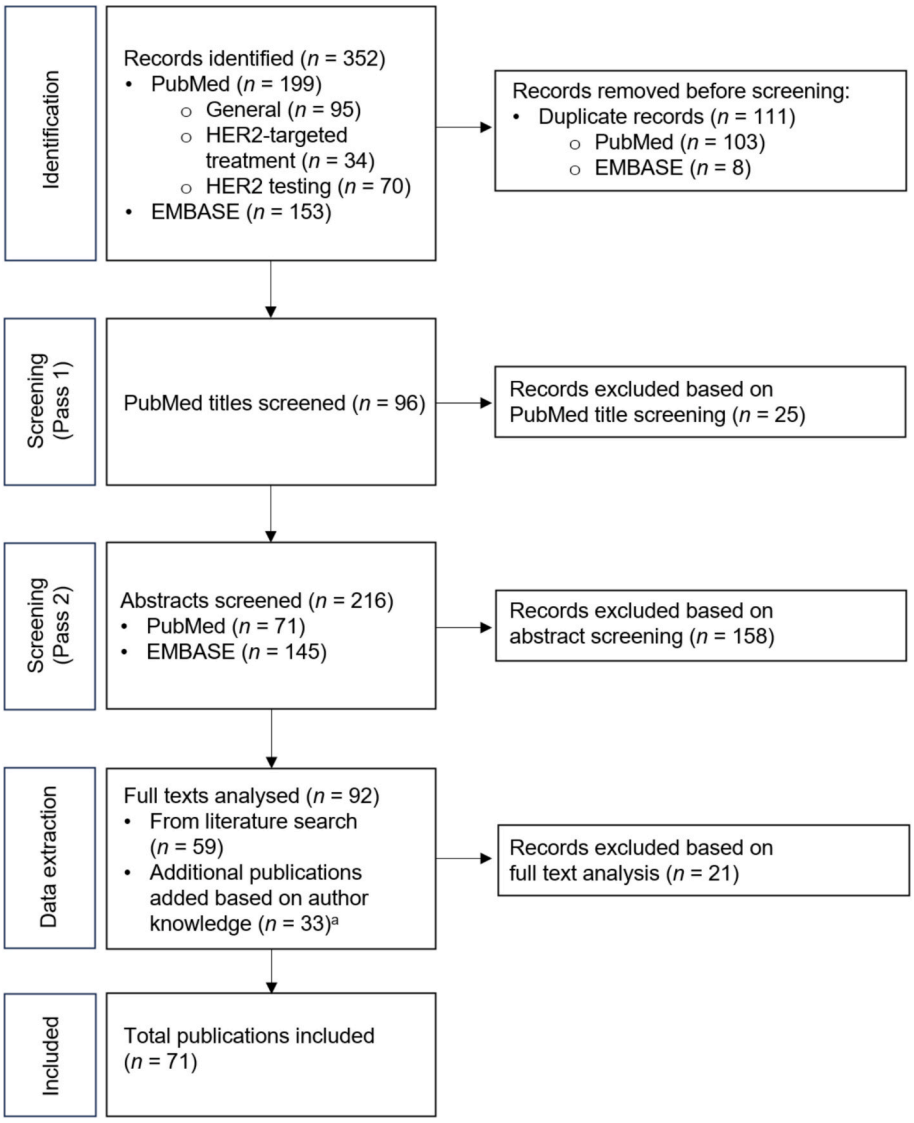

Fig. 1. PRISMA flow diagram of literature search results. HER2, human epidermal growth factor receptor 2; mCRC, metastatic colorectal cancer; PRISMA, Preferred Reporting Items for Systematic Reviews and Meta-Analyses. [a]Thirty-three additional publications were included in the full-text analysis step based on author awareness of key publications in HER2+ mCRC and in cases where a more recent full publication was available to replace congress abstracts identified in the EMBASE congress search [2,18,19,28,30,31,34,46–48,50–54,56–60,67–69,73,76,78,90,98–100,104,107,108].

other gene alterations and treatment response. While some evidence suggests that *HER2* point mutations may be predictive of resistance to anti-epidermal growth factor receptor (EGFR) treatments [34], other studies show no association [35], and further investigation is needed to elucidate potential therapeutic opportunities relating to *HER2* mutations in mCRC.

Testing for HER2 overexpression in mCRC

Testing for HER2 overexpression is not performed routinely for patients with mCRC [36]. The 2025 ESMO Living Guidelines recommend testing via IHC or fluorescence *in situ* hybridisation (FISH) for HER2 overexpression/amplification among patients with *RAS*wt mCRC [2,37], and the National Comprehensive Cancer Network® (NCCN®) recommends HER2 testing via IHC and *in situ* hybridisation (ISH; according to the HERACLES criteria, described in the 'IHC and ISH' section below) or next-generation sequencing (NGS) for suspected or proven metastatic colon adenocarcinoma or rectal cancer with suspected or proven distant metastases [38,39]. Therefore, there is a need for increased awareness and application of HER2 testing for patients with mCRC. However, compared with HER2+ breast and gastric cancer, the methodology and criteria for determining HER2 overexpression in mCRC have not been validated in phase III trials. As such, there has been a lack of consistency in HER2 testing criteria across key clinical trials in HER2+ mCRC (Table 2), and various testing methodologies and criteria are applied in clinical practice across institutions. In the following section, we summarise key publications on HER2 testing in mCRC, and highlight key differences between criteria assessed in mCRC with those employed in breast and gastric cancer.

IHC and ISH

In 2015, Valtorta and colleagues reported their validation study in archival-test (n = 256) and clinical-validation (n = 830) cohorts, which aimed to establish criteria for HER2 overexpression in mCRC to support enrolment of patients into the phase II HERACLES trial. They defined HER2 overexpression/amplification in mCRC as intense (IHC 3+) circumferential, basolateral, or lateral staining in ≥ 50% of cells, or in > 10% to < 50% of cells if accompanied by a positive ISH result (*ERBB2: CEP17* ratio ≥ 2 in ≥ 50% of cells). Equivocal results were defined as moderate (IHC 2+) circumferential, basolateral, or lateral staining in ≥ 50% of cells and were considered to show overexpression/amplification

Table 1
Key findings from identified studies relating to HER2 overexpression and *HER2* mutations in mCRC.

	Study design	Key findings
Overview of HER2 overexpression in mCRC		
Frequency	Prospective IHC analysis of 1,046 primary CRC tumours	HER2 overexpression in CRC overall: 4% [26]
	Systematic literature review and meta-analysis of 52 publications on HER2+ mCRC using an FDA-approved assay	HER2 overexpression in CRC overall: 4% [31]
	Retrospective analysis of 264 patients enrolled in phase II/III trials of the German Rectal Cancer Study Group	HER2 overexpression in patients with rectal cancer: 27% [27]
	Open-label phase II HERACLES trial of 914 patients with *KRAS* exon 2 wt mCRC	HER2 overexpression in patients with *KRAS*wt mCRC: 5% [29]
	Retrospective analysis of 59 patients with *RAS/BRAF*wt mCRC undergoing systemic treatment	HER2 overexpression in patients with *RAS/BRAF*wt mCRC: 7% [30]
Associated clinical characteristics	Systematic literature review and meta-analysis of 52 publications on HER2 + mCRC using an FDA-approved assay	• Rate of HER2 overexpression was statistically higher in *RAS*wt CRC (6%) versus *RAS* mutant CRC (1%); P < 0.0001) [31] • Enrichment of HER2+ CRC among patients with microsatellite stable and left-sided CRC [31]
	Retrospective analysis of 1,645 primary colorectal carcinoma cases	HER2 overexpression correlated with: • Higher disease stage per Union for International Cancer Control classification (P = 0.017) [109] • Presence of lymph node metastases (P = 0.029) [109]
	Retrospective analysis of 3,045 patients with colon cancer enrolled in the PETACC3 adjuvant chemotherapy trial	HER2 overexpression correlated with: • Distal tumour location (i.e. splenic flexure, descending colon, and sigmoid colon) [28,110]
Prognostic impact	Retrospective analysis of the FIRE-1 phase III trial among 208 mCRC tumour samples	No association observed between HER2 overexpression and PFS or OS (P = 0.75 and 0.57, respectively) [32]
	Retrospective analysis of tumour samples from 505 patients with mCRC using NGS and NanoString data	*HER2* amplification was associated with longer PFS (P = 0.018) but not OS (P = 0.13) [33]
Overview of *HER2* mutations in mCRC		
Frequency	Retrospective analysis of 5,786 Chinese patients with CRC	• Overall occurrence of *HER2* alterations: 9% • Occurrence of pathogenic mutations: 5% • Occurrence of CNV: 3% • Occurrence of single nucleotide variants: 6% • Patients with single nucleotide variants had a significantly higher tumour mutation burden than those with CNV (P < 0.0001) [111]
	Systematic literature review of 31 publications related to the predictive value of *ERBB2* point mutations in patients with mCRC	Occurrence of *HER2* point mutations in mCRC: Up to 3% [34]
	Comprehensive genomic profiling study of 8,887 cases of CRC	• 189 *HER2* short variants within 178 mCRC samples, of which 180 (95.8%) were known activating alterations • Most *HER2* short variants encoded missense alterations though there was one case of an exon 20 insertion and two cases of exon 16 deletions [107]
	Retrospective analysis of 419 patients with CRC	Activating *HER2* variants appear to include V842I, S310F, and V777L [108]
Association with other gene alterations	Comprehensive genomic profiling study of 8,887 cases of CRC	Association between *HER2* short variant alterations and a higher occurrence of mutations in *PI3K*, mismatch repair, and Wnt pathways [107]
	Retrospective analysis of 419 patients with CRC	Association between *HER2* mutations and microsatellite instability and *PIK3CA* mutations but no association with *TP53*, *APC*, *KRAS*, *NRAS*, or *BRAF* mutations in tissue [108]
	NGS analysis of 1,300 cases of CRC	No association between mismatch repair deficiency and *HER2* amplification [46]
Association with treatment response	Systematic literature review of 31 publications related to the predictive value of *ERBB2* point mutations in patients with mCRC	• Association between *HER2* point mutations and resistance to anti-EGFR treatments, particularly L755S and R784G • No association between *HER2* mutation and tumour response to HER2-targeted treatments [34]
	Retrospective analysis of 216 patients with *RAS*wt unresectable liver-limited mCRC treated with cetuximab plus chemotherapy or chemotherapy alone	• *HER2* mutations overall had no impact on survival compared with *HER2*wt disease in patients treated with cetuximab plus chemotherapy or chemotherapy alone [35]

CNV, copy number variants; CRC, colorectal cancer; EGFR, epidermal growth factor receptor; FDA, US Food and Drug Administration; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; mCRC, metastatic colorectal cancer; NGS, next-generation sequencing; OS, overall survival; PFS, progression-free survival; wt, wild-type.

if accompanied by a positive ISH result [40]. Compared with the HER2 testing criteria in breast and gastric cancer, the HERACLES criteria require a higher proportion of tumour cells to be stained (≥ 50% vs. ≥ 10%) and allow a broader range of staining patterns (Fig. 2). The ISH positivity criteria are similar across the HERACLES, breast, and gastric criteria (Fig. 2).

The use of FISH for equivocal cases is important considering the presence of tumour heterogeneity in HER2+ mCRC. Of 1,614 patients assessed in a retrospective analysis of the HORIZON III trial, 2% of samples showed HER2 overexpression/amplification; of which, 1% were IHC 3+ and 1% were IHC 2+/ISH+ . The authors noted that IHC 2+ tumours showed more heterogeneity, with cells that weakly expressed HER2 mixed with cells that lacked HER2 expression [36]. An analysis published in 2021 suggested that the use of FISH should be expanded and always used in parallel with IHC for testing of patients with mCRC. The authors performed a concordance analysis of IHC and FISH for identifying HER2 overexpression/amplification in mCRC, and observed an overall concordance rate of 92%. Of note, they observed two cases where patients with a negative IHC sample (0/1+) had a positive FISH result and one patient with an equivocal IHC result (2+) who had a positive FISH result [41]. These findings are supported by a similar retrospective concordance analysis of 49 cases of CRC, in which three cases showed low levels of *HER2* amplification by FISH yet showed negative (0+) staining per IHC and no copy number gains by NGS. The authors proposed the inclusion of FISH in all HER2 testing to ensure that such cases are not missed [42]. Intratumoural heterogeneity of mCRC tumours can be either genetic (a mixture of HER2+ cells with HER2– cells) or nongenetic (a mixture of HER2+ cells with *HER2*-amplified

Table 2
Key clinical trials that have assessed HER2-targeted treatments for patients with HER2+ mCRC.

Trial name	Drug regimen	Trial study design	Patient eligibility criteria	HER2 testing criteria	Efficacy outcomes	Safety outcomes
HERACLES [29,63]	Lapatinib + trastuzumab (N = 27)	Phase II, multicentre, open-label study	*KRAS* exon 2 (codons 12 and 13) wt/HER2+ mCRC Progression while on treatment or within 6 months of treatment with approved standard therapies for mCRC (fluoropyrimidines, oxaliplatin, irinotecan, cetuximab, or panitumumab-containing regimens; with or without anti-angiogenic regimens)	IHC 3 + in > 50% of cells OR IHC 2+ and a *HER2/CEP17* ratio > 2.0 in > 50% of cells by FISH	*ORR, %* Primary analysis: 30 (14–50) Long-term follow-up: 28 (NR) *DoR (95% CI), weeks* Primary analysis: 38 (24–94+) *DCR (95% CI), %* Primary analysis: 59 (39–78) Long-term follow-up: 69 (NR) *Median PFS (95% CI), weeks* Primary analysis: 21 (16–32) Long-term follow-up: 4.7 (3.7–6.1) *Median post-hoc OS (95% CI), weeks* Primary analysis: 46 (33–68) Long-term follow-up: 10.0 (7.9–15.8)	*Most common TRAEs:* Diarrhoea (78%), rash (48%), fatigue (48%), paronychia (33%), and conjunctivitis (19%) *Grade 4/5 TRAEs*: 0
MOUNTAINEER [66,67,70]	Tucatinib + trastuzumab (Primary analysis, N = 84; final analysis, N = 86) OR Tucatinib monotherapy (N = 30)	Phase II, multicentre, open-label study	*RAS*wt/HER2+ unresectable or mCRC Chemotherapy-refractory Previously treated with fluoropyrimidines, oxaliplatin, irinotecan, an anti-VEGF monoclonal antibody, and an anti–PD-L1 or anti–PD-1 monoclonal antibody if the tumour had mismatch repair-deficient proteins or had high microsatellite instability Previous use of anti-EGFR antibodies was not required No previous HER2-targeted treatment permitted	IHC 3+ OR IHC 2 + and amplification by FISH/CISH OR Positivity confirmed by NGS	<u>Primary analysis:</u> *ORR (95% CI), %* Tucatinib + trastuzumab: 38 (28–49) Tucatinib monotherapy: 3 (0–17) *DCR, %* Tucatinib + trastuzumab: 71 Tucatinib monotherapy: 80 <u>Final analysis:</u> *Median DoR per BICR (95% CI), months* Tucatinib + trastuzumab: 15.2 (8.9–20.5) *Median PFS per BICR (95% CI), months* Tucatinib +	<u>Tucatinib + trastuzumab (final analysis)</u> *Most common TEAEs*: Diarrhoea (66%), fatigue (44%), nausea (35%) *Most common grade 3 TEAE:* Hypertension (7%) *TEAEs that led to treatment discontinuation*: 6% <u>Tucatinib monotherapy (primary analysis)</u> *Most common TEAEs:* Diarrhoea (33%), abdominal pain (20%), fatigue (20%) *Most common grade ≥ 3 TEAEs*: ALT/AST increase (7% for both)

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Table 2 (*continued*)

Trial name	Drug regimen	Trial study design	Patient eligibility criteria	HER2 testing criteria	Efficacy outcomes	Safety outcomes
					trastuzumab: 8.1 (4.2–10.2) *Median OS (95% CI), months* Tucatinib + trastuzumab: 23.9 (18.7–28.3)	
MyPathway [85,86]	Pertuzumab + trastuzumab (N = 57)	Phase IIa, multicentre, nonrandomised, open-label study	Targetable, pathway-activating, molecular alterations in the HER2 pathway Previously received standard first-line therapy for mCRC, and in the judgement of the treating physician, had no further therapies suitable to convey clinical benefit No previous HER2-targeted treatment permitted	FISH/CISH: *HER2/CEP17* ratio > 2.0 or *HER2* copy number > 6.0 OR NGS (FoundationOne): Copy number increased OR IHC: 3+	*ORR (95% CI), %* 32 (20–45) *CBR (95% CI), %* 44 (31–58) *Median DoR (95% CI), months* 5.9 (2.8–11.1) *Median PFS (95% CI), months* 2.9 (1.4–5.3) *Median OS (95% CI), months* 11.5 (7.7–NE)	*TEAEs:* 53/57 (93%) *Serious TEAEs:* 10/57 (18%) *Grade 3/4 TEAEs:* 21/57 (37%) *Most common TEAEs:* Diarrhoea (33%), fatigue (32%), nausea (30%) *Most common grade 3/4 TEAEs:* Hypokalaemia (5%), abdominal pain (5%)
TRIUMPH [88]	Pertuzumab + trastuzumab (N = 30)	Phase II, multicentre, single-arm, open-label study	*RAS*wt/HER2 + mCRC Refractory or intolerant to fluoropyrimidine, irinotecan, and oxaliplatin, and to an anti-EGFR antibody No previous HER2-targeted treatment permitted	Tissue: IHC 3 + in > 10% of tumour cells or FISH positive (*HER2/CEP17* ratio ≥ 2.0) OR Positivity confirmed by ctDNA analysis with Guardant360	*ORR (95% CI), %* Tissue-positive patients: 30 (14–50) ctDNA-positive patients: 28 (12–49) *Median DoR (95% CI), months* Tissue-positive patients: 12.1 (2.8–NR) ctDNA-positive patients: 8.1 (2.8–NR) *Median PFS (95% CI), months* Tissue-positive patients: 4.0 (1.4–5.6) ctDNA-positive patients: 3.1 (1.4–5.6) *Median OS (95% CI), months* Tissue-positive patients: 10.1 (4.5–16.5) ctDNA-positive patients: 8.8 (4.3–12.9)	*TRAEs*: 24/30 (80%) *Grade 3 TRAEs*: 3/10 (10%) *Most common TRAEs:* IRR reactions (47%), diarrhoea (37%), stomatitis (13%), malaise (10%)
HERACLES-B [93]	Pertuzumab + T-DM1 (N = 31)	Phase II, multicentre, open-label study	*RAS* (*KRAS* exons 2, 3, 4; *NRAS* exons 2, 3, 4) wt/HER2+ mCRC Progression while on treatment or within 6 months from treatment with approved standard drugs for mCRC (fluoropyrimidine- oxaliplatin-, or irinotecan-containing regimens, with or without anti-	IHC 3 + in > 50% of cells OR IHC 2+ and a *HER2/CEP17* ratio > 2.0 in > 50% of cells by FISH	*ORR (95% CI), %* 10 (0–28) *DCR, %* 77 *Median PFS (95% CI),*	*TRAEs grade ≤ 2:* 84% of cycles *Grade 3 TEAEs:* 2 patients *Most common TEAEs:* Fatigue (18%), hyperbilirubinaemia (9%), thrombocytopaenia (8%), pruritus (8%), nausea/

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Table 2 (*continued*)

Trial name	Drug regimen	Trial study design	Patient eligibility criteria	HER2 testing criteria	Efficacy outcomes	Safety outcomes
			angiogenic or anti-EGFR antibodies)		*months* 4.1 (3.6–5.9)	vomiting (8%), muscular pain (8%)
DESTINY-CRC01 [97,99]	T-DXd Cohort A (N = 53) Cohort B (N = 15) Cohort C (N = 18)	Phase II, multicentre, open-label study	*RAS/BRAF*wt/HER2+ mCRC Prior receipt of and progression on at least two previous treatment regimens, including fluoropyrimidines, irinotecan, oxaliplatin, and anti-EGFR antibodies or anti-VEGF antibodies Prior HER2-antibody or anti-HER2 agent use (except T-DXd) was permitted but a washout period of at least 4 weeks was required	Cohort A: IHC 3+ or IHC 2+ and ISH+ Cohort B: IHC 2+ and ISH– Cohort C: IHC 1+	Final analysis *ORR (95% CI), %* Cohort A: 45 (32–60) Cohort B: 0 (0–22) Cohort C: 0 (0–19) *DCR (95% CI), %* Cohort A: 83 (70–92) Cohort B: 60 (32–84) Cohort C: 22 (6–48) *DoR (95% CI), months* Cohort A: 7.0 (5.8–9.5) Cohort B: NE (NE–NE) Cohort C: NE (NE–NE) *Median PFS (95% CI), months* Cohort A: 6.9 (4.1–8.7) Cohort B: 2.1 (1.4–4.1) Cohort C: 1.4 (1.3–2.1) *Median OS (95% CI), months* Cohort A: 15.5 (8.8–20.8) Cohort B: 7.3 (3.0–NE) Cohort C: 7.7 (2.2–13.9)	Final analysis (Cohorts A, B, C) *TEAEs:* 100%, 100%, 100% *TRAEs:* 96%, 100%, 83% *Grade ≥ 3 TEAEs:* 66%, 47%, 78% *Grade ≥ 3 TRAEs:* 55%, 27%, 50% *Most common grade ≥ 3 TEAEs (across all cohorts):* Decreased neutrophil count (22%), anaemia (14%) Across all cohorts, there were three cases (3.5%) of grade 5 treatment-related ILD/pneumonitis
DESTINY-CRC02 [100]	T-DXd 5.4 mg/kg (N = 82) OR T-DXd 6.4 mg/kg (N = 40)	Phase II, multicentre study	*RAS*wt or mutant HER2+ mCRC Prior receipt of standard therapy Prior anti-HER2 therapy was permitted	IHC 3+ OR IHC 2+ and ISH+	*ORR (95% CI), %* 5.4 mg/kg: 38 (27–49) 6.4 mg/kg: 28 (15–44) *Median DoR by BICR (95% CI), months* 5.4 mg/kg: 5.5 (4.2–8.1) 6.4 mg/kg: 5.5 (3.7–NE) *Median PFS (95% CI), months* 5.4 mg/kg: 5.8 (4.6–7.0) 6.4 mg/kg: 5.5 (4.2–7.0)	*Grade ≥ 3 TRAEs:* 5.4 mg/kg: 41% 6.4 mg/kg: 49% *ILD:* 5.4 mg/kg: 8% 6.4 mg/kg: 13%

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Table 2 (*continued*)

Trial name	Drug regimen	Trial study design	Patient eligibility criteria	HER2 testing criteria	Efficacy outcomes	Safety outcomes
					Median OS (95% CI), months 5.4 mg/kg: 13.4 (12.5–16.8) 6.4 mg/kg: NE (9.9–NE)	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; BICR, blinded independent central review; CBR, clinical benefit rate; CI, confidence interval; CISH, chromogenic *in situ* hybridisation; ctDNA, circulating tumour DNA; DCR, disease control rate; DoR, duration of response; EGFR, epidermal growth factor receptor; FISH, fluorescence *in situ* hybridisation; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ILD, interstitial lung disease; IRR, infusion-related reaction; ISH, *in situ* hybridisation; mCRC, metastatic colorectal cancer; NGS, next-generation sequencing; NE, not estimable; NR, not reached; ORR, objective response rate; OS, overall survival; PD-1, programmed cell death protein 1; PD-L1, programmed cell death ligand 1; PFS, progression-free survival; T-DM1, trastuzumab emtansine; T-DXd, trastuzumab deruxtecan; TEAE, treatment-emergent adverse event; TRAE, treatment-related adverse event; VEGF, vascular endothelial growth factor; wt, wild type.

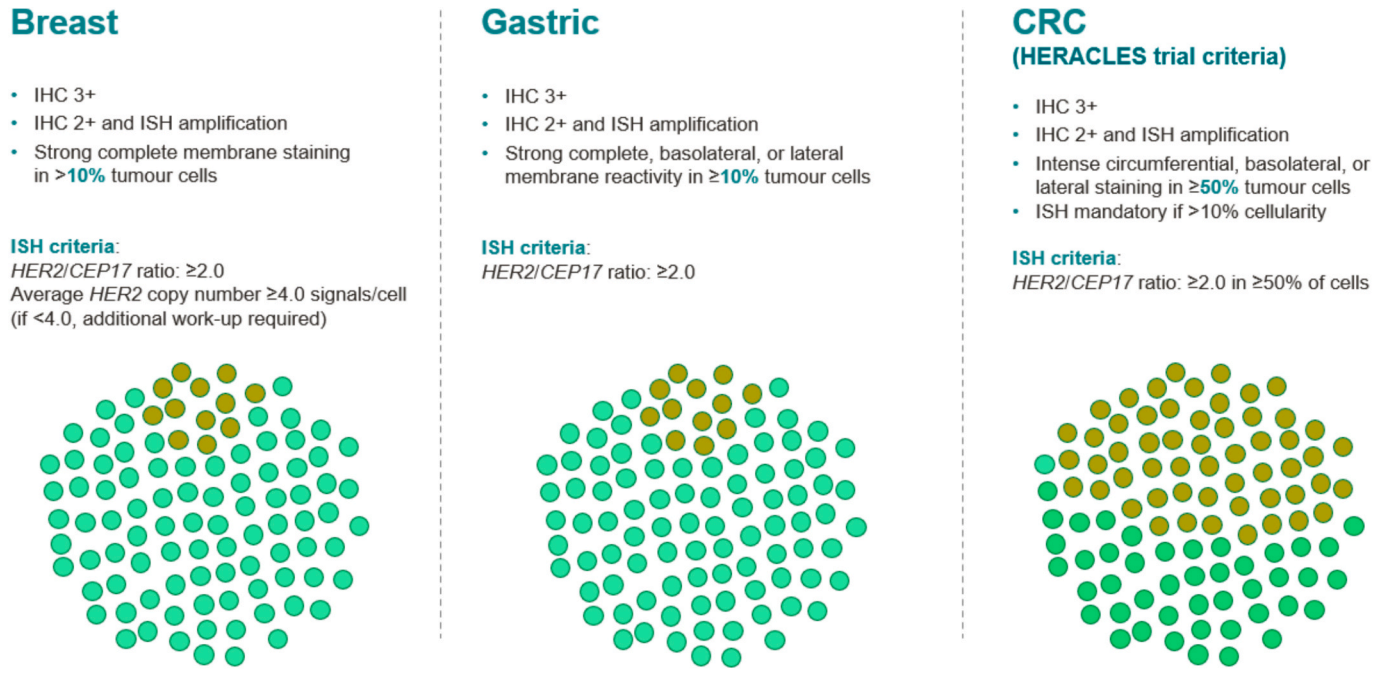

Fig. 2. Differences in HER2 testing criteria between the HERACLES trial for HER2+ mCRC and established criteria used in HER2+ breast and gastric cancer [18,19,40]. CEP17, chromosome 17; CRC, colorectal cancer; IHC, immunohistochemistry; ISH, *in situ* hybridisation; HER2, human epidermal growth factor receptor 2; mCRC, metastatic colorectal cancer. Note: All circles represent tumour cells; light green circles represent cells without IHC staining for HER2 expression and brown circles indicate cells with IHC staining for HER2 expression. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

cells without HER2 protein expression). One study assessed the accuracy of a gene-protein assay for diagnosing intratumoural heterogeneity in mCRC and found that 22% of HER2+ samples displayed intratumoural heterogeneity; this was more commonly seen in IHC 2+ samples (60%) compared with IHC 3+ samples (16%; P = 0.06) [43].

A concordance analysis published in 2023 compared the use of the breast and gastric HER2 testing criteria for use in mCRC based on patient samples from the phase III MOUNTAINEER trial. In MOUNTAINEER, patients were enrolled if they demonstrated HER2 overexpression/amplification in at least one of the following methods: IHC, ISH, and NGS. In this analysis, samples were submitted for local HER2 testing in addition to scoring by the breast and gastric criteria. The authors observed 100% concordance in HER2 status between the breast and gastric criteria, and 99% concordance with local HER2 IHC score. The authors concluded that their results support the use of the breast or gastric criteria for diagnosing HER2+ mCRC until there is an available validated HER2 assay for mCRC [44].

Tissue-based NGS

NGS has also been investigated for its potential use as a diagnostic tool for identifying *HER2* amplification in mCRC. A retrospective analysis published in 2017 investigated the utility of comprehensive genomic sequencing in detecting *HER2* amplification compared with historical methodology (IHC and FISH). Of 201 patients with CRC, 10 patients (5%) were found to be HER2+ (on both IHC/ISH and comprehensive genomic sequencing), and the HER2 testing result was the same for all patients using both IHC/ISH and comprehensive genomic sequencing [45]. These findings are supported by a similar study, in which *HER2* amplification detected by NGS was found to predict HER2 overexpression per IHC in samples from patients with CRC; although, the authors noted that IHC was required to capture protein-level heterogeneity [46].

In 2020, Fujii and colleagues developed harmonised diagnostic criteria for identifying HER2 overexpression/amplification in mCRC: IHC 3+ or 2+ and a FISH *ERBB2/CEP17* ratio of ≥ 2.0, with tumour content > 10% for surgically resected specimens and necessary tumour

content not defined for biopsy specimens. The authors then validated the criteria against copy number variants (CNV) as assessed by NGS. For patients with HER2+ or HER2– disease, gene copy number (determined by FISH) correlated with CNV (determined by NGS; r = 0.98; P < 0.0001). Among all patients who met the consensus criteria, CNV was > 4.0. The authors concluded that their criteria demonstrated consistency between IHC/FISH and CNV determined by NGS [47].

Circulating tumour DNA-based NGS

In addition to tissue-based NGS, NGS using plasma samples from a liquid biopsy has been investigated as a surrogate diagnostic marker in mCRC. In 2019, Siravegna and colleagues assessed the sensitivity of plasma copy number for detecting *HER2* amplification in mCRC, and investigated the relationship between cell-free circulating tumour DNA (ctDNA) copy number and response to HER2-targeted therapy using HER2+ samples from patients enrolled in the HERACLES-A trial. The authors observed a 98% sensitivity rate, with 46/47 evaluable samples identified as HER2+ by cell-free ctDNA copy number. It is of note that an adjusted HER2 plasma copy number of ≥ 25.82 was identified to correlate with best overall response and progression-free survival (PFS; P = 0.03) [48].

Real-world testing for HER2 overexpression in mCRC

We identified several publications that investigated the reality of HER2 testing for mCRC in clinical practice, including initiatives to improve education of healthcare professionals (HCPs) treating patients with mCRC and understanding the potential cost implications of increased testing. Based on the favourable outcomes of the HERACLES and MyPathway studies, one US institution began reflexively testing all cases of invasive or mCRC for HER2 overexpression/amplification using IHC and ISH according to the HERACLES criteria. They observed a real-world incidence of HER2+ mCRC aligned with clinical trial experience and retrospective studies (4% of patients). The authors stated that if a laboratory is capable of testing for HER2 overexpression among other tumour types, it is feasible to apply this testing for mCRC, which might provide broader treatment options for patients [49]. To improve HCP awareness and knowledge of HER2 testing in CRC, one international group of authors launched an online accredited education program (GetSMART), comprising four learning modules with an embedded knowledge/confidence-based assessment. Of 159 HCPs, the confidence percentage in identifying *HER2* aberrations significantly increased from the start to the end of the program (16% vs. 29%, P < 0.001). Similarly, of 116 HCPs, the performance in identifying *HER2* aberrations significantly increased (41% vs. 68%, P < 0.001) [50]. While many studies have demonstrated the potential value of comprehensive genomic profiling for identifying *HER2* amplification in patients with mCRC, the use of such methodology is not routinely employed across many global regions. One US study assessed the cost impact of replacing 20% of usual testing methods with comprehensive genomic profiling using a decision analytic model based on a hypothetical US health-plan setting. Based on an expected number of 1,112 incident cases of mCRC per year, they identified 521 cases of missed opportunities for genomics-driven treatment (442 due to lack of testing and 79 due to lack of comprehensive genomic profiling). Further, they observed only a minimal budget impact associated with broadened use of comprehensive genomic profiling [51]. As HER2 overexpression is rare in patients with other activating mutations such as *KRAS* and *BRAF* mutations [46], a possibility in areas with limited resource would be to consider HER2 testing for *KRAS/BRAF*wt cases only.

HER2-targeted treatment options for mCRC

For patients with left-sided unresectable *RAS*wt mCRC, chemotherapy combined with anti-EGFR therapies (cetuximab or panitumumab) forms the basis of treatment options in the first-line setting [2]. However, there is significant evidence that supports the role of HER2 overexpression in the development of resistance to anti-EGFR therapies, which was first identified based on preclinical evidence in 2011 and later clinical findings in 2019 [52–54]. In a meta-analysis of five retrospective cohort studies (comprising 594 patients with *RAS*wt mCRC treated with anti-EGFR regimens) published in 2023, HER2+ status was associated with a 2.84 × higher risk of death or progression (PFS) compared with HER2– status. There were no statistically significant differences in overall survival (OS) [55]. Another study investigated the predictive value of EGFR and its dimerisation partner for response to treatment with cetuximab in patients with *RAS*wt mCRC. They found that patients with IHC 3+ staining intensity for HER2 and HER3 had a shorter PFS [56].

With poor outcomes for patients with HER2+/*RAS*wt mCRC who develop resistance to anti-EGFR therapies, there is an important unmet need for novel targeted treatments in this patient group. In 2011, Bertotti and colleagues reported their preclinical findings, in which they identified HER2 as an effective therapeutic target in cetuximab-resistant CRC based on patient-derived xenografts. They observed that dual inhibition of HER2 and EGFR resulted in long-lasting tumour regression [52]. These findings were supported by further research by Leto and colleagues in 2015, in which they analysed the effect of HER2-targeted treatments (trastuzumab, lapatinib, and afatinib) on tumour growth using patient-derived tumour grafts and cell-line xenografts. They found that dual vertical blockade with trastuzumab + lapatinib produced a greater antitumour response than either of these treatments alone [57]. The underlying mechanism for this observation is a synergistic relationship between trastuzumab and lapatinib, whereby lapatinib-induced accumulation of HER2 at the tumour cell surface potentiates trastuzumab-dependent cell cytotoxicity [58,59]. In a later preclinical investigation by Kulukian et al, tucatinib exhibited antitumour activity that was enhanced by combination with trastuzumab (including a patient-derived colorectal xenograft model); tucatinib also demonstrated increased selectivity for HER2 compared with lapatinib or neratinib in tumour cell lines [60].

Clinical data on HER2-targeted treatments in mCRC

Since the publication of the aforementioned pivotal preclinical studies in HER2+ mCRC [52,57–60], there has been a growing body of evidence that supports the use of HER2-targeted treatments in this patient population and highlights the value of early HER2 testing at diagnosis of mCRC. Recent systematic literature reviews and meta-analyses have demonstrated favourable efficacy and safety outcomes for HER2-targeted therapies for the treatment of HER2+ mCRC [61,62]. The ESMO Living Guidelines recommend HER2-targeted treatment (trastuzumab + tucatinib or trastuzumab deruxtecan [T-DXd]) from the third-line setting for HER2+ mCRC [37]. The NCCN Guidelines® recommend HER2-targeted treatment (trastuzumab + [pertuzumab, lapatinib, or tucatinib] for IHC 3+, IHC 2+/ISH+, or NGS-amplified HER2+ *RAS/BRAF*wt disease and T-DXd for IHC 3+ HER2+ disease) from the second-line setting, or in the first-line setting for *RAS/BRAF*wt disease when intensive therapy is not recommended (trastuzumab + [pertuzumab or lapatinib or tucatinib] only) [38,39].

Based on the publications identified by our systematic literature review, in this section, we provide an overview of data on the use of HER2-targeted treatments in patients with HER2+ mCRC according to drug class. In Table 2, we summarise key data across pivotal clinical trials for HER2+ mCRC.

Tyrosine kinase inhibitors

Lapatinib + trastuzumab. Following the preclinical findings of Bertotti et al and Leto et al regarding dual inhibition with lapatinib + trastuzumab based on patient-derived xenografts [52,57], the phase II HERACLES trial was a proof-of-concept study of patients with *KRAS exon 2* wt/HER2+ mCRC who received dual-targeted therapy with lapatinib +

trastuzumab. Among 27 evaluable patients, there was an objective response rate (ORR) of 30%, median PFS of 21 weeks, median OS (post hoc) of 46 weeks, and 22% of patients experienced grade 3 adverse events (AEs) [29]. In an updated analysis of HERACLES after 6.7 years of follow-up, antitumour activity had been sustained (with an ORR of 28% among 32 evaluable patients). Median PFS was 4.7 months and median OS was 10.0 months. The authors reported disease progression in the central nervous system in 6/32 evaluable patients, which suggests that patients with HER2+ mCRC may have a propensity for developing brain metastases, as is observed for HER2+ breast and gastric cancer [63].

Lapatinib and trastuzumab are not currently approved by the FDA and EMA for the treatment of HER2+ mCRC [10,11,64,65].

Tucatinib + trastuzumab. In the primary analysis of the phase II open-label MOUNTAINEER trial (median follow-up: 16.3 months), treatment with tucatinib + trastuzumab was associated with an ORR of 38% among 84 evaluable patients. Median PFS per blinded independent central review was 8.2 months and median OS was 24.1 months. Grade 3/4 AEs were observed in 38% of patients. These findings led to the tucatinib combination regimen becoming the first FDA-approved HER2-targeted treatment for chemotherapy-refractory HER2+/*RAS*wt mCRC [66]. In the final analysis of MOUNTAINEER (median follow-up: 32.4 months), the median duration of response, PFS, and OS for tucatinib + trastuzumab were 15.2 months, 8.1 months, and 23.9 months, respectively [67]. An exploratory analysis of MOUNTAINEER found that tucatinib + trastuzumab had broad clinical activity in both the overall study population and subgroups of patients with concomitant clinically relevant genomic alterations (including *PIK3CA*, *APC*, and *TP53* alterations). Potential factors associated with greater clinical benefit included earlier lines of treatment (second line and below), left-sided primary tumours, higher gene copy number, and increased *HER2/CEN17* ratio. NGS analysis using ctDNA found *RAS* and *HER2* mutations did not predict primary treatment resistance; common acquired alterations identified at time of progression included *PIK3CA*, *RTKs*, and *KRAS/NRAS*, and loss of *HER2* amplification was rare [68]. In MOUNTAINEER, HER2 overexpression/amplification was determined centrally by IHC, ISH, and/or NGS testing; a retrospective analysis observed that HER2 status confirmed by any of the three methods predicted treatment response to tucatinib + trastuzumab [69]. As predicted by preclinical studies of single versus dual HER2 blockade, the tucatinib monotherapy arm of MOUNTAINEER was associated with an ORR of 3% among 30 patients. These findings highlight the value of dual blockade with tucatinib + trastuzumab in HER2+ mCRC [57,70]. Patient-reported outcomes for the MOUNTAINEER trial demonstrated sustained health-related quality of life for 65 patients treated with either tucatinib monotherapy or in combination with trastuzumab, evidenced by stable mean changes in the European Organization for the Research and Treatment of Cancer Quality of Life Questionnaire score from baseline through to the end of treatment [71].

Additional tucatinib studies are ongoing to investigate different treatment combinations and lines of treatment. The efficacy and safety of tucatinib + trastuzumab + fluorouracil, leucovorin, and oxaliplatin (FOLFOX) among patients with HER2+ gastrointestinal cancers (including an mCRC cohort) is being assessed in the ongoing open-label phase Ib/II SGNTUC-024 trial [72]. The ongoing phase III MOUNTAINEER-03 trial will assess PFS as a primary endpoint, and OS and confirmed ORR as secondary endpoints for tucatinib + trastuzumab + modified FOLFOX6 as a first-line treatment regimen versus standard-of-care treatment for patients with HER2+/*RAS*wt locally advanced unresectable or mCRC [73].

Tucatinib is currently approved under accelerated approval by the FDA for the treatment of adult patients with HER2+/*RAS*wt unresectable or mCRC that has progressed following treatment with fluoropyrimidine-, oxaliplatin-, and irinotecan-based chemotherapy [74]; tucatinib is not currently approved by the EMA for use in HER2+ mCRC [75].

Pyrotinib + trastuzumab. A phase IIa study (HER2-FUSCC-G) of pyrotinib + trastuzumab observed a higher ORR among patients with *RAS*wt/HER2+ disease (57%) compared with patients with HER2+ mCRC overall (50%). Patients with *RAS/BRAF*wt disease also had prolonged PFS (7.53 vs. 1.63 months) and OS (not reached vs. 4.13 months) compared with *RAS/BRAF* mutant disease. The overall rate of grade 3 treatment-emergent AEs was 31% [76]. In a similar, single-arm, open-label phase II trial, pyrotinib + trastuzumab was associated with an ORR of 22% among 18 evaluable patients with HER2+ mCRC and 33% among a subset of 12 patients who had *RAS*wt disease. Median PFS was 3.4 months for the overall population and 4.3 months among patients with *RAS*wt disease; OS was not reached. The proportion of patients with grade 3 AEs was 75% [77]. Another exploratory single-arm phase II trial of pyrotinib + trastuzumab in heavily pre-treated patients with HER2+ mCRC observed an ORR of 23.5%, disease control rate of 88.2%, median PFS of 6.2 months, and median OS of 21.1 months, with improved outcomes for patients with *RAS/BRAF*wt disease. Grade 3 treatment-related AEs occurred in 30% of patients (all diarrhoea) [78].

Pyrotinib is not currently approved by the FDA or EMA for use in HER2+ mCRC.

Neratinib + cetuximab. In the phase Ib NSABP FC-7 trial of patients with quadruple wt (*KRAS*, *NRAS*, *BRAF*, *PIC3KA*) mCRC, neratinib + cetuximab was not associated with any objective responses. Of 21 enrolled patients, only two patients had *HER2* amplification at the start of the study; although, this increased to four patients following trial enrolment and receipt of anti-EGFR treatment [79]. The phase II NSABP FC-11 trial of neratinib + cetuximab planned to include a cohort of patients who had received prior anti-EGFR treatment and had either *HER2* amplification or a *HER2* mutation; however, this arm closed due to poor accrual (n = 4) [80].

Neratinib is not currently approved by the FDA or EMA for use in HER2+ mCRC [81,82]. Cetuximab is approved by the FDA and EMA for use in EGFR-expressing, *RAS*wt mCRC [83,84].

Monoclonal antibodies

Pertuzumab + trastuzumab. The phase IIa basket study, MyPathway, investigated the use of pertuzumab + trastuzumab in patients with advanced solid tumours harbouring potentially actionable molecular alterations. Among the subset of patients with HER2+ mCRC (n = 57), there was an ORR of 32%, median PFS of 2.9 months, and median OS of 11.5 months. Grade ≥ 3 treatment-emergent AEs were observed in 37% of patients [85,86]. A noninterventional study compared OS outcomes from patients with HER2+ mCRC receiving pertuzumab + trastuzumab in MyPathway with a real-world patient cohort receiving routine clinical care, and observed an estimated hazard ratio for OS of 0.73 in favour of pertuzumab + trastuzumab (95% confidence interval 0.18–3.90) [87].

TRIUMPH was a phase II trial that aimed to assess ORR as a primary endpoint for pertuzumab + trastuzumab among patients with HER2+ mCRC prospectively confirmed by tissue- or ctDNA-based analysis. The study met its primary endpoint, with comparable ORR rates for patients with HER2 overexpression/amplification confirmed by tissue (30%) versus ctDNA analysis (28%), which were vastly improved compared with a matched real-world reference population treated with standard-of-care salvage therapy (0%). Median PFS and OS were 4.0 months and 10.1 months, respectively, based on tissue analysis, and were 3.1 months and 8.8 months, respectively, based on ctDNA analysis. Grade 3 treatment-related AEs were observed in 10% of patients. These results confirmed the applicability of ctDNA-based testing for *HER2* amplification in mCRC and for monitoring response to treatment [88].

In the phase II TAPUR trial, clinical outcomes for pertuzumab + trastuzumab were assessed for 28 patients with HER2 overexpression.

The results for the primary study endpoint of disease control were favourable (ORR of 25% and disease control rate of 54%); median PFS was 17.2 weeks and median OS was 60.0 weeks [89].

The phase II SWOG-S1613 trial compared efficacy outcomes for patients with HER2+/*RAS*wt mCRC who received HER2-targeted treatment (pertuzumab + trastuzumab) with anti-EGFR therapy and chemotherapy (cetuximab + irinotecan). Among 54 included patients, there was no difference in median PFS (4.7 months for pertuzumab + trastuzumab vs. 3.7 months for cetuximab + irinotecan). Median OS was not reached for pertuzumab + trastuzumab and was 24.7 months with cetuximab + irinotecan. The ORR was 34.6% for pertuzumab + trastuzumab and 28.0% for cetuximab + irinotecan. The rate of grade ≥ 3 AEs was lower for pertuzumab + trastuzumab (23%) compared with cetuximab + irinotecan (46%) [90].

Pertuzumab is not currently approved by the FDA or EMA for use in HER2+ mCRC [91,92].

Antibodydrug conjugates

Trastuzumab emtansine + pertuzumab. The single-arm phase II HERACLES-B trial assessed clinical outcomes for trastuzumab emtansine (T-DM1) + pertuzumab among 31 evaluable patients with *RAS/BRAF*wt/HER2+ mCRC. Although the study did not meet its primary endpoint of ORR (with a rate of 10%), the rate of stable disease was 68% and median PFS was comparable with other HER2-targeted treatments (4.1 months). Two patients experienced treatment-related grade 3 AEs [93]. The potential utility of T-DM1 in patients with HER2+ mCRC at progression to treatment with lapatinib + trastuzumab was planned to be assessed in the phase II HERACLES RESCUE trial. However, owing to poor accrual the study was terminated prematurely [94].

T-DM1 is not approved by the FDA or EMA for use in HER2+ mCRC [95,96].

Trastuzumab deruxtecan. In the open-label phase II DESTINY-CRC01 trial, trastuzumab deruxtecan (T-DXd) was investigated as a monotherapy for the treatment of patients with HER2+ mCRC (cohort A). Among 53 patients, the ORR was 45%, median PFS was 6.9 months, and median OS was not reached [97]. In exploratory biomarker analyses of the DESTINY-CRC01 trial, response to treatment was found to be associated with higher tumour or plasma HER2 expression, and was also observed in patients with and without *RAS* or *PIK3CA* mutations [98]. In the final analysis of the DESTINY-CRC01 trial, including long-term efficacy and safety data, the ORR for cohort A was 45%, with a median PFS of 6.9 months and median OS of 15.5 months. The occurrence of grade ≥ 3 AEs in cohort A was 66%. The authors noted the potential safety risk of T-DXd in relation to interstitial lung disease/pneumonitis, which was associated with three treatment-related deaths across all cohorts [99]. This led to the use of a lower dose in the subsequent DESTINY-CRC02 trial.

The phase II DESTINY-CRC02 trial investigated the clinical efficacy and safety of T-DXd at both the 5.4 mg/kg and 6.4 mg/kg doses among patients with HER2+ mCRC. In the primary analysis, for the 5.4 mg/kg and 6.4 mg/kg doses, the confirmed ORR was 38% and 28%; median PFS was 5.8 and 5.5 months; and median OS was 13.4 months and not estimable, respectively. Grade ≥ 3 treatment-related AEs were observed in 41% and 49% of patients in the 5.4 mg/kg and 6.4 mg/kg arms, respectively [100].

T-DXd is currently approved under accelerated approval by the FDA for use in patients with unresectable or metastatic HER2+ (IHC 3+) solid tumours who have received prior systemic treatment and have no satisfactory alternative treatment options [101]; T-DXd is not approved by the EMA for use in HER2+ mCRC [102].

Real-world evidence on HER2-targeted treatments in mCRC

We identified three studies relating to the use of HER2-targeted therapies in the real-world setting. In one study, the authors applied a Bayesian design to assess longitudinal efficacy for lapatinib + trastuzumab among patients who were treated in clinical practice and had similar characteristics to the HERACLES-A trial population. Lapatinib + trastuzumab was confirmed to have a favourable safety profile and be clinically effective, with a likelihood of response of 26% [103]. In relation to testing, a real-world survey of gastrointestinal oncologists across a range of different global countries found HER2 is not typically included in routine testing and there was significant regional variation in treatment patterns in relation to tumour sidedness [104]. Despite an extensive and growing collection of clinical trial data on the utility of HER2-targeted treatments for HER2+ mCRC and their inclusion in clinical guidelines, a retrospective real-world analysis of treatment patterns in the US, published in 2023, found that many patients still do not receive HER2-targeted therapy and have heterogenous treatment patterns. The authors concluded that there is a need for greater awareness of treatment options for this patient population [105].

Author guidance on optimal testing and treatment of HER2+ mCRC

Considering the lack of certainty surrounding optimal HER2 testing and the broad range of HER2-targeted treatments under investigation, we have proposed a testing and treatment algorithm based on the evidence summarised from our literature search and clinical experience across the countries represented within the author group (Fig. 3).

In the future, potential treatment strategies may include earlier use of HER2-targeted treatment (for example, the use of first-line tucatinib following up to two rounds of modified FOLFOX6 per the MOUNTAINEER-03 trial regimen [73,106]). However, the utility of HER2-targeted treatments in the first-line setting is still under investigation and will depend on the outcome of ongoing trials.

Conclusions

In the era of precision oncology, it is essential to test every patient for molecular biomarkers that may provide meaningful therapeutic benefit, particularly for patient subgroups with a high unmet need such as HER2+ mCRC. HER2 overexpression occurs in a clinically relevant number of patients with mCRC (5% in *RAS*wt mCRC [29]) and has shown consistent therapeutic actionability with various regimens. Many trials have shown the benefit of HER2-targeted treatments for the management of patients with *RAS*wt/HER2+ mCRC, including combinations of monoclonal antibodies (e.g. trastuzumab + pertuzumab), and the strongest evidence emerging from trials using double vertical blockade with an anti-HER2 tyrosine kinase inhibitor + trastuzumab (e.g. MOUNTAINEER, HERACLES) or with an antibody–drug conjugate (e.g. DESTINY-CRC01 and 02). However, in the real world, patients are not being routinely tested for HER2 overexpression and so are unable to receive HER2-targeted treatments. Many studies have explored and validated different testing methods and criteria for identifying HER2 overexpression in patients with mCRC,; although there remains an important unmet need for consolidated diagnostic criteria as seen in HER2+ breast and gastric cancer. Despite the ongoing challenges for the testing and treatment of HER2+ mCRC, our findings highlight the importance of early testing for HER2 overexpression at diagnosis of mCRC, including a fresh biopsy before initiating HER2-targeted treatment, and awareness of emerging clinical trial data to ensure that this patient population can receive the most suitable treatment.

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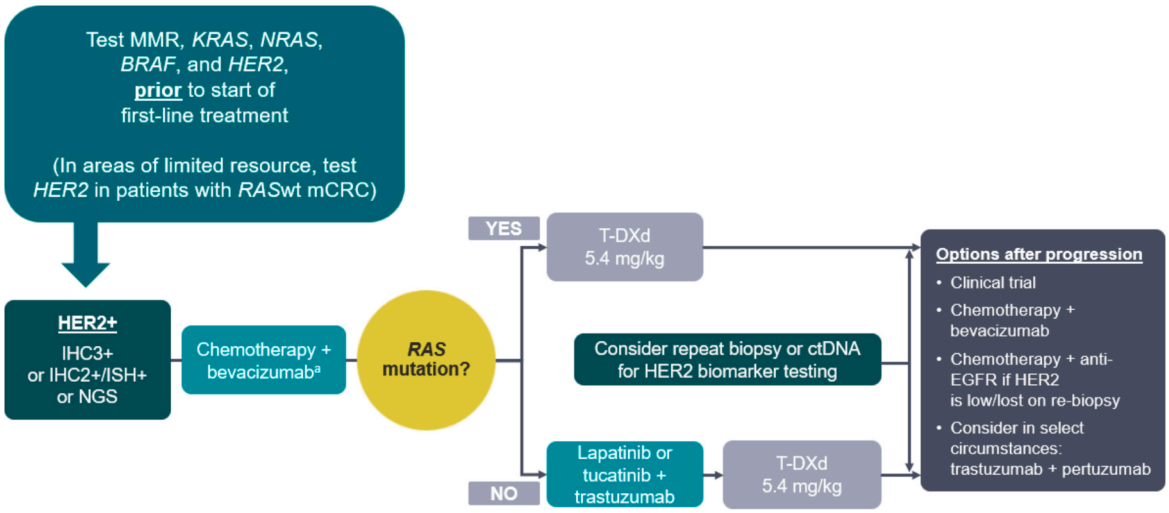

Fig. 3. Proposed testing and treatment algorithm for HER2+ mCRC based on evidence from the literature search and the authors' clinical experience. ctDNA, circulating tumour DNA; FDA, US Food and Drug Administration; FOLFOX, fluorouracil, leucovorin, and oxaliplatin; EGFR, epidermal growth factor receptor; EMA, European Medicines Agency; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridisation; mCRC, metastatic colorectal cancer; MMR, mismatch repair; NGS, next-generation sequencing; T-DXd, trastuzumab deruxtecan; wt, wild-type. [a]Consider enrolling patients in a clinical trial of HER2-targeted therapy; the ongoing MOUNTAINEER-03 trial is assessing the efficacy of tucatinib + trastuzumab + modified FOLFOX6 as first-line treatment versus standard of care in patients with HER2+ RASwt mCRC and may provide a potential first-line HER2-targeted treatment option in the future. Note: This guidance is based on author opinion and includes treatment options that have not received FDA or EMA approval.

Declaration of competing interest

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Appendix A. Supplementary data

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Glossary

AE: adverse event
ASCO: American Society of Clinical Oncology
ALT: alanine aminotransferase
AST: aspartate aminotransferase
BICR: blinded independent central review
CBR: clinical benefit rate
CI: confidence interval
CISH: chromogenic *in situ* hybridisation
CNV: copy number variants
CRC: colorectal cancer
ctDNA: circulating tumour DNA
DCR: disease control rate
DoR: duration of response
EGFR: epidermal growth factor receptor
EMA: European Medicines Agency
ERBB2: HER2
ESMO: European Society for Medical Oncology
ESMO GI: European Society for Medical Oncology Gastrointestinal Cancers Congress
FDA: US Food and Drug Administration
FISH: fluorescence *in situ* hybridisation
FOLFOX: fluorouracil leucovorin and oxaliplatin
FOLFOX6: tucatinib + trastuzumab + modified
HCP: healthcare professional
HER2: human epidermal growth factor receptor 2
IHC: immunohistochemistry
ILD: interstitial lung disease
IRR: infusion-related reaction
ISH: in situ hybridisation
mCRC: metastatic colorectal cancer
MMR: mismatch repair
NCCN: National Comprehensive Cancer Network
NE: not evaluable
NGS: next-generation sequencing
NR: not reached
ORR: overall response rate
OS: overall survival
PD-1: programmed cell death protein 1
PD-L1: programmed cell death ligand 1
PFS: progression-free survival
PRISMA: Preferred Reporting Items for Systematic reviews and Meta-Analyses
T-DM1: trastuzumab emtansine
T-DXd: trastuzumab deruxtecan
TEAE: treatment-emergent adverse event
TRAE: treatment-related adverse event
USCAP: United States and Canadian Academy of Pathology
VEGF: vascular endothelial growth factor
wt: wild-type