Review

Low and Ultra-low HER2 in Human Breast Cancer: An Effort to Define New Neoplastic Subtypes

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Review

Low and ultra-low HER2 in human breast cancer: an effort to define new neoplastic subtypes

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ABSTRACT: HER2-low and ultra-low breast cancer (BC) have been recently proposed as new subcategories of HER2 BC, re-considering the immunohistochemical negative score of 0, 1+ or score of 2+/in situ hybridization (ISH) negative phenotype. In the present review we have outlined the needed criteria to exactly distinguish HER2 low and ultra-low BC. Recent clinical trials have demonstrated significant clinical benefits of novel HER2 directing antibody-drug conjugates (ADCs) in treating these group sof tumors. In particular, Trastuzumab-deruxtecan (T-Dxd), a HER2-directing ADC has been recently approved by the U.S. Food and Drug Administration as the first targeted therapy to treat HER2-low BC. Furthermore, ongoing trials, such as the DESTINY-Breast06, are currently evaluating ADCs in patients with HER2-ultra low BC. Finally we hope that new guidelines may help to codify HER2 low and ultra-low BC, increasing our knowledge of tumor biology and improving a targetable new therapeutical treatment.

Keywords: HER2 expression; Breast cancer; Neoadjuvant treatment; HER2-low carcinomas; HER2 ultra-low carcinomas

INTRODUCTION

Breast cancer (BC) is worldwide the most frequent cancer diagnosed in women and it continues to be a relevant cause of death[1], even if new strategies in clinical-therapeutic management have been introduced improving the patients' outcomes in overall survival (OS), progression free survival (PFS) and pathology complete response (pCR)[2].

At first, therapeutic strategies for BC were previously guided by the expression of hormonal receptors (HR) and Ki67 proliferation index[3]. Successively, the rising importance of the prognostic role of the anti-human epidermal growth factor receptor 2 (anti-HER2) brought to the establishment of a new classification of BC and the development of new therapeutic strategies based on the molecular characteristics of each tumour[3–5]. This classification includes four categories of BCs with different prognostic and predictive values[4,5]. In detail, Luminal A and Luminal B categories are considered HER2-negative tumours, that express HR at different levels[4,5]. Moreover, Luminal B tumours express low levels of progesterone (PR) and present different genetic mutations, resulting less differentiated and more aggressive tumors compared to Luminal A tumours[6]. The third category is represented by Triple Negative BC (TNBC), which do not express HR nor HER2 and are characterized by a more aggressive behaviour, younger age of incidence, worst prognosis, fewer treatment options, often associated with BRCA1 mutation[7]. Finally, HER2-positive tumours are characterized by the

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overexpression of HER2 due to HER2 gene amplification, aggressive behaviour and poor prognosis[4]. On this way, the assessment of HER2 status by using immunochemistry (IHC) and in situ hybridization (ISH) represents a mandatory parameter for a correct characterization and therapeutic choice for BC patients. The American Society of Clinical Oncology/ College of American Pathologists (ASCO/CAP) recommendations to define HER2 status, with its last update in 2018[8], subdivides HER2 IHC results in two categories: HER2-negative and HER2-positive, assigning a specific score to each sample, based on the intensity and the pattern of expression in the cell membrane as well as the percentage of positive cells[9]. HER2-negative category includes tumours with score 0, 1+, and 2+ without HER2 gene amplification on ISH, whereas HER2-positive category includes tumours with score 3+ and 2+ with HER2 gene amplification on ISH[8].

The demonstration of BC cases with HER2 amplification raised the question to develop specific monoclonal antibodies against HER2 to realize a specific target therapy[10]. On this way, specific therapeutic agents, such as trastuzumab[11,12], pertuzumab[13,14], lapatinib[15–17], have been developed, representing thus an extremely important tool in the precision era medicine, able to significantly improve the life expectation of HER2-positive BC patients[18]. In fact, HER2 overexpression is present approximately in 20% of patients with BC[19] and it is usually associated with more aggressive behaviour[20], hight risk of disease recurrence and a shorter OS[20–22] in comparison to HER2-negative (HER2-) BCs. To date, no effective target treatments are approved for HER2-negative BC patients since several studies demonstrated a beneficial effect of the addition of trastuzumab to chemotherapy only in tumours with HER2 overexpression[23–25].

However, recently the introduction of novel anti-HER2 antibody-drug conjugates (ADC) strategies showed promising response rates and progression-free survival (PFS) also in the so-called HER2-"low" BCs, a new definition attributed to tumours which present a HER2 IHC score of 1+ or 2+ without HER2 amplification[26–28]. Furthermore, it has been raised evidence concerning the benefit of the treatment response also in HER2-"ultra-low" BCs, characterised by a HER2 IHC score 0[29].

In the present review, we have focused on current applicative aspects concerning BC management of patients with the new codified HER2-low and ultra-low status.

How to currently establish HER2 status

The first problem to define HER2 overexpression in BC patients has been considered how to measure the presence of HER in breast cancer cells. Although HER2 gene amplification has been correctly identified by fluorescence in situ hybridization (FISH) or in situ hybridization (ISH)[30], the immunohistochemical demonstration of the corresponding HER2 oncoprotein expression has been considered the most practical approach in laboratories[31] (Table 1).

Table 1. Interpretation of HER2 IHC staining and ISH analysis.

	HER2 score 3+	HER2 score 2+	HER2 score 1+	HER2 score 0
IHC	Complete, intense and circumferential membrane staining in >10% of tumor cells	Incomplete and/or weak to moderate circumferential membrane staining in >10% of the tumor cells **or** presence of intense, complete and circumferential membrane	Incomplete or faint/barely perceptible membrane staining in >10% of tumor cells	Incomplete or faint/barely perceptible membrane staining in ≤ 10% of tumor cells No staining

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		staining in ≤10% of the tumor cells		
ISH	**AMPLIFIED** HER2/CEP17 ratio was ≥ 2.0 or HER2/CEP17 ratio < 2.0 with average HER2 copy number ≥ 6.0	**NOT AMPLIFIED** HER2/CEP17 ratio < 2.0 with average HER2 copy number < 4.0		
	HER2 POSITIVE	**HER2 LOW**	**HER2 ULTRA LOW**	
			HER2 NEGATIVE	

The ASCO/CAP guidelines for HER2 interpretation were first introduced in 2007[32] stating that HER2 status should be initially assessed by IHC and subsequently confirmed by FISH in case of equivocal results[33]. The 2007 guidelines set a cutoff of 30% positive neoplastic cells for HER2 IHC and the HER2 gene resulted amplified if HER2/chromosome enumeration probe 17 (CEP17) ratio was > 2.2 or HER2 copy number was > 6.0 in FISH analysis[33]. In 2013, the revised ASCO/CAP guidelines[34] focused on further decreasing false-negative results that could affect the recruitment of patients who could effectively benefit from a target therapy. Positive score (3+) was defined by the presence of a complete, intense and circumferential membrane staining in >10% of tumor cells, while equivocal score (2+) was defined by the presence of incomplete and/or weak to moderate circumferential membrane in >10% of the invasive tumor cells or the presence of intense, complete and circumferential membrane staining in ≤10% of the invasive tumor cells. Otherwise, negative scores (1+ and 0) were defined by the presence of incomplete and weak membrane staining in >10% of the invasive tumor cells or the absence of staining or a barely perceptible staining in < 10% of tumor cells, respectively.

FISH analysis has been sometimes considered as the first choice to determine the HER2 status, but more frequently it has been applied to verify equivocal score (2+) as an amplified HER2 gene[34]. BC cases resulted amplified when HER2/CEP17 ratio was ≥ 2.0 or HER2/CEP17 ratio < 2.0 with average HER2 copy number ≥ 6.0[34]. Regarding ISH analysis, largely applied in cases of equivocal IHC results (score 2+), the last ASCO/CAP update takes in consideration five different ISH patterns[8]. HER2 resulted amplified in group 1 (HER2/CEP17 ratio ≥ 2.0 and average HER2 copy number ≥ 4.0), not amplified in group 5 (HER2/CEP17 ratio < 2.0 with average HER2 copy number < 4.0) and equivocal in group 2 (HER2/CEP17 ratio ≥ 2.0 and average HER2 copy number < 4.0), group 3 (HER2/CEP17 ratio < 2.0 and average HER2 copy number ≥ 6.0) and group 4 (HER2/CEP17 ratio < 2.0 and average HER2 copy number ≥ 4.0 and < 6.0)[8]. For these last three groups, it is crucial to include a concomitant rigorous IHC check concerning all embedded neoplastic material to achieve the most accurate evaluation of HER2 status. Finally, regarding groups 2 and 4, if HER2 IHC is 2+, then the ISH result is marked as not amplified, whereas in the group 3, if HER2 IHC is confirmed 2+, the ISH result is marked as amplified[35,36].

In clinical practice, these guidelines have been used to discriminate HER2-negative BCs from HER2-positive BCs, the latter eligible for target therapy with anti-HER2 monoclonal antibodies, such as Trastuzumab, Pertuzumab and Margetuximab, able to improve the clinical outcome of these tumors[23–25,37–39]. However, only 20% of BCs result HER2-positive, leaving the rest with fewer treatment options, confined to endocrine therapy and/or chemotherapy with less clinical benefits and more side effects[40–42].

Nevertheless, HER2 status interpretation, and subsequently an accurate classification of BCs patients, can be affected by several factors, being HER2 heterogeneity the most important one as elsewhere documented in other neoplasms[33,43,44]. In fact, intratumoral HER2 heterogeneity has been

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observed in various type of cancers including a subset of BCs[36]. Moreover, several studies have shown that HER2 heterogeneity can affect the accurate HER2 status assessment and it has been reported to be more common in HER2 equivocal status (2+) with an incidence of 16-36% HER2-positive BCs [45,46]. Different HER2 heterogeneous patterns can be found in BCs, as small clusters of amplified cells intermingled to clusters of not-amplified cells or they can appear as distinguished fields of amplified and not-amplified cells[36,45]. Furthermore, HER2 heterogeneity can be found not only inside the primary tumor, but also between primary BCs and metastases or between primary tumor and recurrent or metachronous lesions[33,47,48]. In the light of these evidences, some studies have reported that HER2 heterogeneity has been associated with poor response to trastuzumab and poor clinical outcome in HER2-positive primary and metastatic BCs[35,46,49]. In addition, HER2 heterogeneity has been associated with lower disease-free survival rates in HR-positive BCs as well as with incomplete response to neoadjuvant therapy[50]. Furthermore, several studies evaluating the concordance and/or discordance rate of HER2 IHC and ISH, showed a small percentage of cases (approximately 8-9%)[51], in which BCs were classified as HER2-negative IHC (score 0 and 1+), resulted instead amplified on ISH[51–56]. The main causes of this discrepancy include technical issues, such as samples handling and fixation, interpretation bias, intratumoral HER2 heterogeneity, but also the evidence of BCs with low HER2 amplification (HER2/CEP ratio ≥ 2 with average HER2 signals/cell 4-6)[51]. These cases can potentially benefit from a HER2-target therapy, but actually according ASCO/CAP guidelines[8], their IHC interpretation do not consider an ISH evaluation, excluding thus these patients from a proper targeted treatment.

IDENTIFICATION AND DEFINITION OF HER2-LOW BC

Whitin the large cohort of HER2-negative BC there is a significant heterogeneity, with distinct biological features, especially in tumors IHC score 1+ or 2+ but ISH not amplified, and IHC score 0[57,58]. These tumors HER2- IHC 1+/2+, ISH not amplified, have been recently defined as "HER2 low-masked" or simply "HER2 low-positive", representing about the 45-55% of HER2-negative BC[26,29,57,58]. This new proposed subtype, although not official, has been considered "equivocal", since it has got some characteristics similar to HER2+ BC, but actually it doesn't appear targetable with standard anti HER2 drugs[12]. Consequently, in a specific trial it no clinical benefit has been achieved by adding only Trastuzumab to adjuvant chemotherapy in high risk invasive HER2-low BC[12]. Therefore, monoclonal antibodies appear to be a not appropriate choice in HER2-low BC, since their activity basically consist in blocking the aberrant HER2 signalling (via dimerization inhibition) and the antibody-dependent cellular cytotoxicity[59]. However, the real paradigm shift in the HER-2 low BC field is related to the use of new anti-HER-2 antibody conjugated drugs (ADCs), whose good results in progression free survival (PFS) and response rate (RR) have been shown in several studies[60–62] and deeply demonstrated by the results of the DESTINY-Breast 03 trial. This evidence, summarised at the ESMO 2021 congress[63] and furtherly underlined by DESTINY-Breast 04 trial, revealed significant benefits of ADCs use in patients with metastatic HER-2 low BC[64,65]. Consequently, in August 2022, the US Food and Drug Administration approved Trastuzumab-Deruxtecan for the treatment of patients with HER2-low metastatic BC, adding this new treatment in National Comprehensive Cancer Network Guidelines for this subgroup of patients[64–67].

On a molecular perspective, HER2-low BCs are associated with several mutation, such as PIK3CA (31%), GATA3 (18%), TP53 (17%) and ERBB2 (8%) with an higher prevalence of FGFR1 amplification (defined as ≥10 copy number gain)[68]. Moreover, HR status seems to associate to the HER2-low expression in BC, being more common in tumours with HR expression (65%) rather than triple negative BCs (TNBC) (37%)[60].

The evaluation of HER2-low status has not been formally defined, as no specific procedures have yet been established[26]. The details about the score concern either weak to moderate complete membrane staining observed in more than > 10 of tumour cells (IHC 2 + equivocal not amplified), either incomplete membrane staining that is faint/barely perceptible and in more than 10% of tumour cells (IHC 1+). Unfortunately, the assessment of HER2-low status using conventional testing techniques

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may present elements inaccuracy and therefore, it may occur that reproducibility evaluations of HER2 testing in HER2 low cases have revealed temporal as well as interobserver heterogeneity[2]. To help standardise protocols, the US FDA recently approved the VENTANA PATHWAY anti-HER2/neu (4B5) rabbit monoclonal primary antibody for IHC assessment of HER2 expression as the first companion diagnostic test; therefore, it was tested and used in the DESTINY 04 trial in metastatic BS patients to identify and select only low HER2 cases[69]. The Ventana system will remain the first, but probably not the only one approved to identify low HER2 patients, as new evidence has showed the Dako system detects HER2 expression with higher sensitivity compared to the Ventana system, not only in ISH-positive BC tumours, but also HER2 tumours without gene amplification (IHC 1+ /2+) thus adding more patients identified as HER-2 low patients and selected for new ADC targeted therapies[70]. On the light of these suggestions, it is now mandatory to better standardise protocols establishing which is the test more appropriate for stratifying HER-2-negative as well as identifying HER-2 low cases[66]. As consequence, to accurately define HER-2 low cases, implement validation and quality control systems may be applied in a reproducible format, recruiting more additional patients potentially eligible for HER2-targeted therapies[66,71]. Artificial intelligence (AI) and machine learning programs have been shown to perform well in terms of speed and accuracy in assessing HER2 status and predicting anti-HER2 treatment response, but specific pathologist training in reading IHC-ISH test results remains the current gold standard[72–74].

On the other hand, the HER2-low status should not to be confused with intratumour heterogeneity phenomenon[75], generally responsible of unexpected positive response to target therapy with monoclonal antibody in HER2-negative BC[75]. Recently, several studies have tried to find the presence of a predictive marker strictly connected with HER2-low patients that would take advantage from the use of monoclonal antibody therapy[67,76–78]. In order to increase the number of HER2-low BC patients suitable to be included in HER2 targeted therapy[71], several studies started to classify all new possible HER2 sub-groups, analyzing clinical and molecular landscape of HER2-low BC, more accurately as possible. Therefore, a new classification has been proposed to apply a more specific targeted therapy for each subcategory either in HER2-low expression[18,29,79].

On this way, many efforts have been performed by clinical trials to evaluate the real efficacy of HER2 target-therapy in low-HER2 expressor patients, focalizing data concerning mismatch repair (MMR) genes/ proteins, and other related biomarkers[80–84]. These evidences are based on very sporadic findings since HER2 activating mutations are described in less than 2% of BC, with a higher frequency in HR-positive BCs in comparison to TNBC and likewise in lobular carcinomas than in ductal ones[81].

IDENTIFICATION AND DEFINITION OF HER2 ULTRA-LOW BC

Although HER2-0 scored BCs are generally considered to inadequately responsively to monoclonal antibody, it has been reported that among these patients exist a cohort to be identified, recently defined as HER2-ultra low[85]. This proposed subtype could represent another subcategory eligible for ADCs target therapies, immunohistochemically characterized by faint/barely perceptible and incomplete staining in <10% of tumour cells, but not amplified on FISH[85]. Ongoing studies, such as the DESTINY-Breast06 trial are currently evaluating ADCs in patients with HER2-ultra low BC, who could benefit from new conjugated HER2-targeted therapies also in the cohort previously scored as 0 HER-2[59]. However, this minimal HER2 ultra-low expression could probably be sufficient for the novel ADCs in order to exert their specific cytotoxic effect on the neoplasm. In this "ultra-low" phenotype activating genetic mutations not related to the IHC status; have been reported, representing an alternative way of activating the HER2 pathway in BC[82]. In detail, V777L ERBB2 mutation is an activating mutation as it strongly increases the phosphorylation of signalling proteins, indicating enhanced activity of the tyrosine kinase[82]; for this reason, BC having neoplastic cell HER2 V777L-mutated can be administrated with TK inhibitors (like lapatinib and neratinib)[82]. Furthermore, MutL deficiency (connected with mismatch repair system alteration) represents a mutation related to endocrine treatment resistance, which appears in 15–17% of

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oestrogen-receptors positive (ER+)/HER2-negative BC[83,86,87]. It may be hypothesized that the loss of MutL expression could activate HER2 without receptor overexpression; consequently, MutL loss has been proposed as a marker to stratify ER+/HER2−negative BC patients probably responsive to anti-HER agents[87]. So, HR+/HER2−negative BC with a molecular mutation with MutL loss show a good response to a combination of anti-HER2 drugs and endocrine treatments[84,88]. Finally, despite molecular characteristics, the very slight HER2 expression shown in the ultra-low HER2 subgroup could also be due to the limitations of pathological testing, in which false negative results could be artefactually related to inadequate formalin fixation process or insufficient sensitivity of the IHC assay[89,90]

NEW TREATMENT OPTIONS IN HER2 LOW E ULTRA-LOW PATIENTS

The development of the novel antibody-drug conjugates (ADCs) has recently revolutionized the therapeutic scenario of HER2 low tumors, demonstrating a survival benefit in this setting (Table 2). Trastuzumab Deruxtecan (T-DXd) is a novel ADC consisting of a monoclonal antibody targeting HER2 and a potent payload, the topoisomerase I inhibitor deruxtecan. These components are linked together with a ratio 1:8 by a tetrapeptide linker. After internalization of T-DXd, deruxtecan is released by lyposomale enzymes and inhibits the topoisomerase I, which in turn leads to the break of double strand DNA[91]. Furthermore, deruxtecan is able to pass the cell membrane and kill tumor cells nearby regardless of HER2 expression. This is called the Bystander-effect[92]. This explains the increased activity of this agent in HER2-low tumors, as compared with older HER2-targeting agents, including previous generation ADCs, such as T-DM1. In a phase I trial, T-DXd showed promising antitumor activity in 54 pretreated HER2 low metastatic breast cancer (MBC) patients with a reported 37% ORR and a median of PFS 11.1 months[93]. These encouraging results were confirmed in a phase III trial, DESTINY Breast 04. In this study, patients with HER2 low MBC, who had received one or two prior lines of chemotherapy, were randomized 2:1 to receive T-DXd or physicians' choice of chemotherapy. The trial demonstrated the superiority of T-DXd over treatment physician choice in terms of both PFS (9.9 vs. 5.1 months, respectively; HR 0.50; p<0.001) and OS (23.4 vs. 16.8 months, respectively; HR 0.64; p=0.001)[67]. A subgroup analysis by hormone receptor (HR) expression showed that the advantage was consistent also in the HR+ group, with a median PFS of 10.1 months in T-DXd cohort vs 5.4 months in the treatment physician's choice arm (HR 0.51, P <0.001) and a median OS of 23.9 months vs 17.5 months, respectively (HR 0.64, p=0.003)[67].

Only a small number of HR-negative patients – i.e. triple negative breast cancer (TNBC) - were included in the trial (11.3% of all the enrolled patients). In an exploratory analysis of this subgroup HR- a median PFS of 8.5 months in T-DxD group vs 2.9 months (HR 0.46) was shown and a median OS of 18.2 months in T-Dxd cohort vs 8.3 months in the treatment physician's choice was reported[67]. Due to these impressive data, T-DXd represents a novel important therapeutic option for HER2 low BC and a further treatment option for HER2 low TNBC.

An ongoing phase 3 study is evaluating the superiority of T-DXd vs chemotherapy in patients with HR+ HER2 low MBC (DESTINY Breast 06, NCT04494425). In addition to single agent use, several studies are investing the combination of novel HER2-targeting ADCs and immune checkpoint inhibitors (ICIs), based on a strong preclinical rationale. Preclinical studies have reported a synergistic activity between T-DXd and ICIs targeting PD-1 and CTLA-4, supporting the clinical development of T-DXd-immunotherapy combinations[94,95]. Preliminary data of the arm 6 of the phase 1/2 BEGONIA trial, evaluating the combination T-DXd with the PD-L1 inhibitor durvalumab in first-line HR-/HER2-low MBC patients, have been recently presented[96].

Durvalumab plus T-DXd showed promising early safety and efficacy in first line HER2-low–expressing TNBC, with a 66.7% ORR, irrespective of PD-L1 expression. T-DXd was also evaluated in combination with the PD-1 inhibitor nivolumab in pretreated MBC in a phase 1b trial (NCT03523572)[97]. The combination was associated with a safety profile consistent with the expected toxicities of each drug and an activity (ORR 65.6% in HER2+ and 50% in HER2-low with a median PFS of 11.6 months and 7.0 months, respectively) in line with single agent T-DXd, questioning the

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additional benefit provided by the combinatorial use. Potential reasons for the differential activity seen with T-DXd plus immunotherapy combinations between first-line and pretreated patients might be attributable to a changed tumor immune microenvironment after multiple lines of treatments.

Other ADCs in development with the same target but distinct payload targeting HER2 low expression are Trastuzumab Duocarmazine (SYD 985) and Disitamab Vedotin (RC48-ADC). Trastuzumab duocarmazine (SYD 985) is a new ADC consisting of the monoclonal antibody trastuzumab, covalently linked via a linker to a DNA-alkylating agent, duocarmazine. In a phase 1 trial that included 49 patients (32 HR+ and 17 HR-) with HER2 low MBC, trastuzumab duocarmazine reported a promising antitumor activity. Interestingly, in the HR+ and HR- cohorts, an ORR of 28% and 40% was reported respectively, with a median of PFS of 4.9 months in HR+ and a median of 4.1 months in HR-[62].

Disitamab vedotin is another novel ADC that combines a novel anti-HER2 antibody, hertuzumab, linked via a cleavable linker to a microtubule inhibitor. Preliminary data on a cohort of 48 patients showed promising tumor activity with an ORR of 40% and a median PFS of 5.7 months, reporting a greater benefit in HER2 2+/ISH negative patients (ORR 42.9%) vs HER2 1+ patients (ORR 30.8%)[98] .

Table 2. Available data of current preclinical and clinical trials of novel ADCs.

Drug	*Population*	*Clinical Trial*	*Result*
Trastuzumab Deruxtecan (T-DXd)	pretreated HER2 low MBC	NCT02564900 (phase I)[93]	ORR = 37% PFS = 11.1 months
Trastuzumab Deruxtecan (T-DXd)	HER2 low MBC pretreated with chemotherapy	NCT03734029 (DESTINY Breast 04 -phase III)[67]	In HR+ patients: PFS = 9.9 months OS = 23.4 months In HR- patients: PFS = 8.5 months OS = 18.2 months
Trastuzumab Deruxtecan (T-DXd)	HR+ HER2 low MBC	NCT04494425 (DESTINY Breast 06 - phase III)	ongoing
T-DXd + anti-PD-L1	Preclinical study[94]		Enhanced antitumour effect by increasing of T-cell activity and upregulation of PD-L1 expression in xenograft mouse models
T-DXd + CTLA-4	Preclinical study[95]		Enhanced antitumour effect by increasing tumour-infiltrating CD4 and CD8
T-Dxd + Durvalumab	HER2 low locally advanced/metastatic TNBC	NCT03742102 (BEGONIA - phase Ib/II)[96]	ORR = 66.7% ongoing
T-Dxd + Nivolumab	Pretreated HER2 low MBC	NCT03523572 (phase Ib)[97]	ORR = 50% PFS = 7 months
Trastuzumab + duocarmazine	Pretreated HER2 low MBC	NCT02277717 (phase I)[62]	In HR+ patients: ORR = 28% PFS = 4.9 months In HR- patients: ORR = 40%

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			PFS = 4.1 months
Hertuzumab + Disitamab Vedotin	Pretreated HER2 low MBC	NCT02881138 NCT03052634 (phase I/Ib)[98]	ORR = 40% PFS = 5.7months

CONCLUSIONS

Recent published data and ongoing clinical trials have defined HER2-low and ultra-low expression in breast, suggesting that new categorization and a new standardized approach to HER2 evaluation in BC is fervently requested. Accepting the new scoring and the consequent sub-grouping the clinical treatment will dramatically revolutionize. Therefore, the acquired knowledge understanding of HER2-low and ultra-low BC has produced further efforts including basic and translational research as well as clinical studies in this newly recognized targetable group of BC. The development of the novel ADCs has recently changed the therapeutic scenario of HER2 BC, demonstrating a survival benefit in these patients. Furthermore, in the precision medicine era, oncologists and pathologists will have the possibility to better define HER2 low and ultra-low BC as separated categories, finally standardized by guidelines and eligible to new therapeutic options.

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