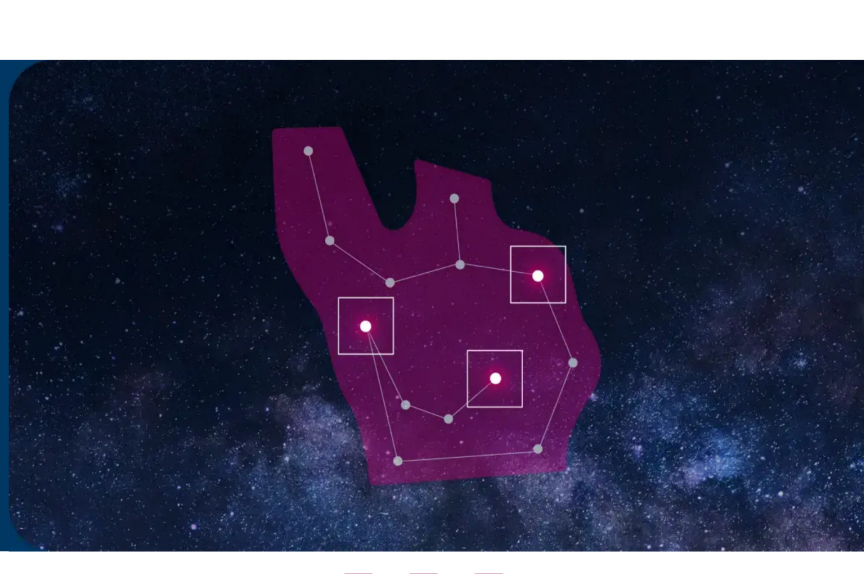
HER2 Expression in Breast Cancer

Explore guideline recommendations and actionable best practices for HER2 IHC testing in breast cancer.



Reading time: **22 min**

Scoring HER2 Expression Across the Full Spectrum Helps to Inform Treatment Decisions[1,2]

Knowing human epidermal growth factor receptor 2 (HER2) status in breast cancer guides care options and informs eligibility for targeted therapies, which is why the NCCN Clinical Practice Guidelines In Oncology (NCCN Guidelines®) recommend testing for this biomarker **in all patients with breast cancer.**[1]

Since 2007, the American Society of Clinical Oncology (ASCO) and College of American Pathologists (CAP) have provided guidance for HER2 immunohistochemistry (IHC) testing.[2] These clinical practice guidelines are periodically updated to meet changing practice needs and incorporate new evidence.

Previously, clinical classification of HER2 status was binary, with assays and best practices optimized to identify HER2-positive (IHC 3+ or IHC 2+/*in situ* hybridization [ISH]+) breast tumors vs HER2-negative.[3] This changed with the **2023 ASCO/CAP guideline update** that offers additional guidance around reporting HER2 IHC 1+, IHC 2+/ISH- results, and best practices for differentiating IHC 1+ and IHC 0 tumors.[2]

ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization; NCCN, National Comprehensive Cancer Network.

HER2 Expression May Inform Prognostic and Predictive Outcomes[4,5]

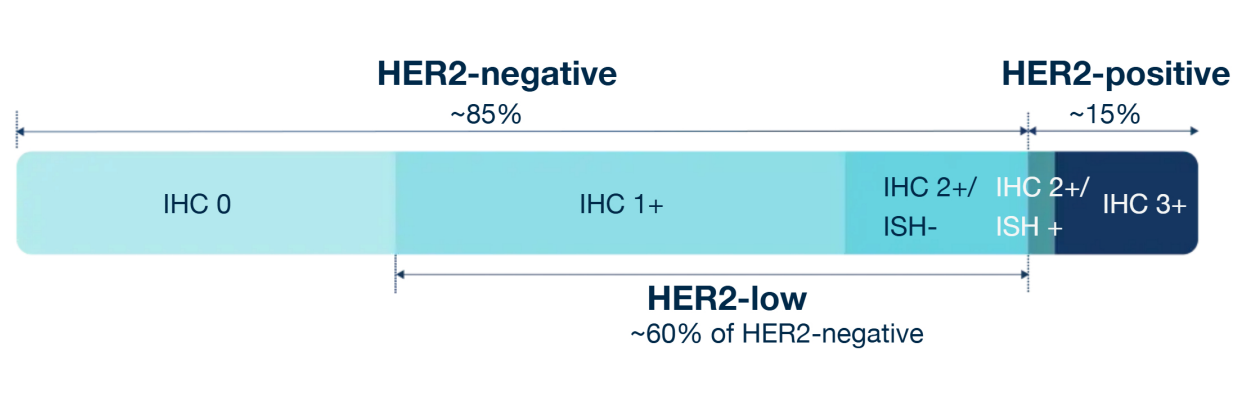
The pathologist's role in HER2 status assessment is critical for targeted treatment

HER2 is overexpressed in approximately 15% of primary breast cancers.[4] HER2 is both prognostic and predictive in breast cancer.[5] When activated, it causes tumor cell proliferation and drives disease progression.[5] Breast cancers are classified by hormone receptor (HR) status (positive or negative; HR+/-) and HER2 status (positive or negative).[6]

The development of HER2-directed therapies has been practice-changing and has significantly improved overall survival in the HER2-positive breast cancer population.[5,7] Consequently, testing for this biomarker has become a routine part of the diagnostic work-up for patients with breast cancer.[5]

HER2, human epidermal growth factor receptor 2; HR, hormone receptor.

Around 6 in 10 Patients Categorized as HER2-Negative Have Discernible Low Levels of HER2 Expression[4,8,9]



Adapted from Tarantino P, et al.; Schettini F, et al. and Rakha EA, et al.

Discrete scoring across the full spectrum of HER2 expression further differentiates each patient[2,4]

As clinical research evolves, it is becoming increasingly relevant to evaluate and differentiate each case of breast cancer with a discrete IHC score.[2,4] This is due to the clinical significance of HER2 extending beyond the binary classification of positive or negative.[4] A subset of HER2-negative expression has now been defined as HER2-low (IHC 1+ or IHC 2+/ISH-).[8,10] Approximately 60% of patients with breast cancer who are categorized as HER2-negative have discernible low levels of HER2 expression.[8] Ongoing research is investigating whether breast cancers scored as HER2 IHC 1+ and IHC 2+/ISH- show biological differences from those scored as IHC 0.[4,8]

The CAP Breast Biomarker Reporting template states that HER2 IHC 1+ and IHC 2+/ISH- may be reported as HER2-low.[11]

CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization.

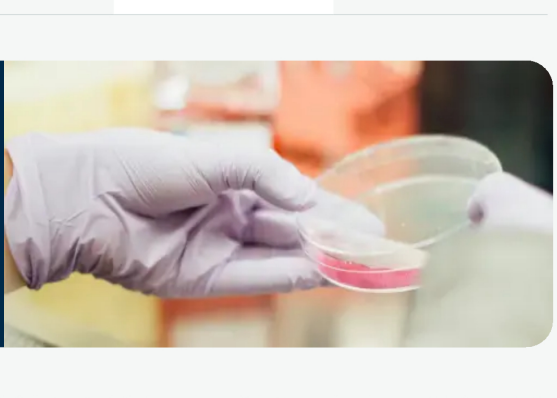
There Are Four FDA-Approved Diagnostic Assays for Measuring HER2 Alterations[12]

ASCO/CAP guidelines recommend the use of FDA-approved assays for HER2 testing in breast cancer.[13]

Assay name (alphabetical order)	Bond™ Oracle™ HER2 IHC System	HercepTest™	InSite™ HER-2/neu	PATHWAY anti-HER2/neu
Platform	BOND-MAX[14]	Dako Automated Link Platform[16]	BioGenex i6000™ Automated Staining System or the OptiMax® Plus Consolidated Staining System[17]	BenchMark ULTRA[18]
Antibody clone/type	CB11[14,15] Mouse monoclonal antibody	RpAb[16] Rabbit polyclonal antibody	CB11[17] Mouse monoclonal antibody	4B5[18] Rabbit monoclonal antibody
Manufacturer	Leica Biosystems[14,15]	Agilent/Dako[18]	BioGenix[17]	Roche/Ventana[18]

See a list of major testing labs in your area.

Find a Testing Lab



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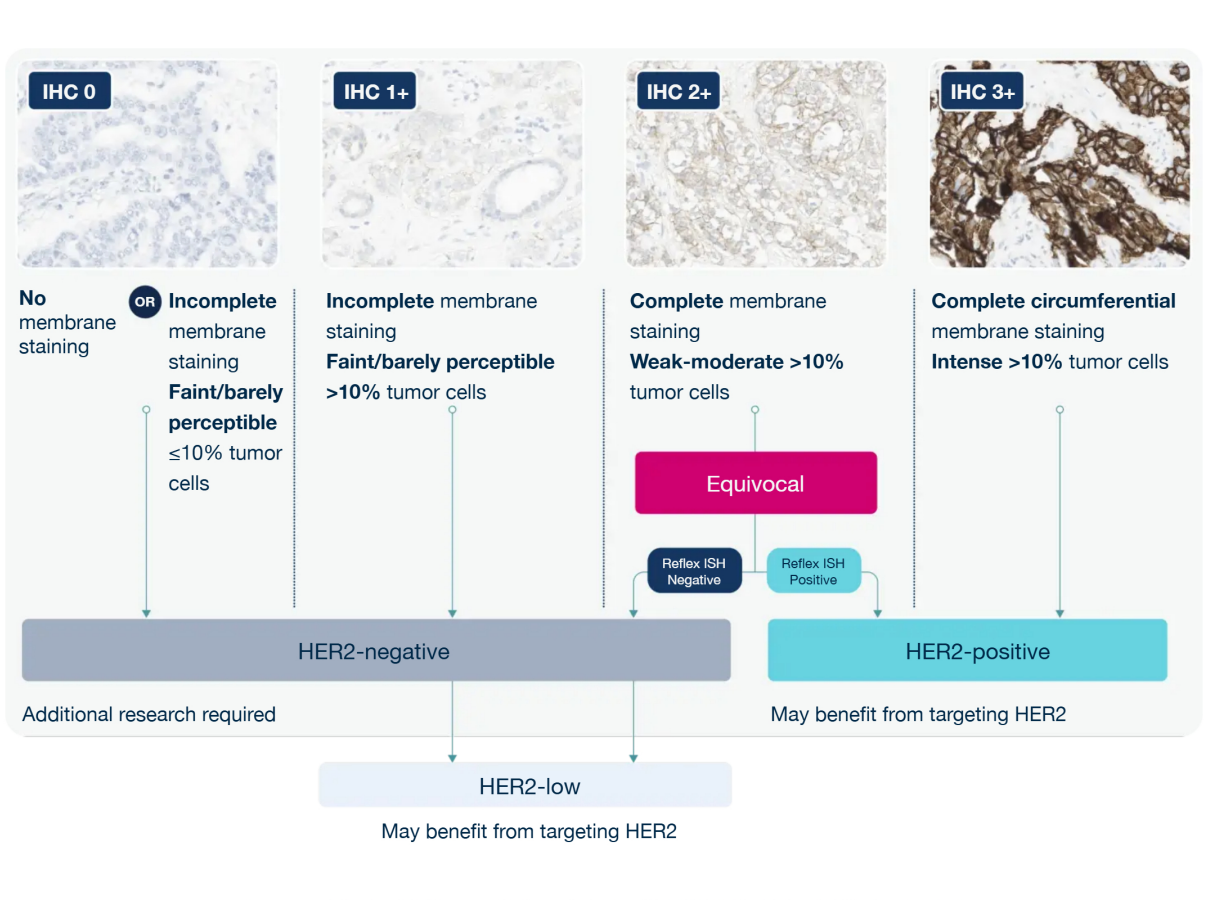
ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; FDA, US Food and Drug Administration; HER2, human epidermal growth factor receptor 2.

How HER2 Results Are Interpreted and Reported Can Have Implications for Patient Care

An alternative ASCO/CAP scoring algorithm to reflect the complete range of HER2 expression[1,2]


IHC 0
IHC 1+
IHC 2+
IHC 3+
No membrane staining
OR
Incomplete membrane staining
Faint/barely perceptible
≤10% tumor cells
Incomplete membrane staining
Faint/barely perceptible
>10% tumor cells
Complete membrane staining
Weak-moderate >10% tumor cells
Complete circumferential membrane staining
Intense >10% tumor cells
Equivocal
Reflex ISH Negative
Reflex ISH Positive
HER2-negative
HER2-positive
Additional research required
May benefit from targeting HER2
HER2-low
May benefit from targeting HER2

Images contributed from HER2Know.com.[19] Figure adapted from 2023 ASCO/CAP guidelines HER2 testing algorithm.[2]

Use controls with a wide range of HER2 expression including IHC 1+ staining to help ensure that the assay has an appropriate limit of detection.[2]

HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization.

Scoring HER2-Low Expression May Be Clinically Relevant for Patients With Breast Cancer[20]

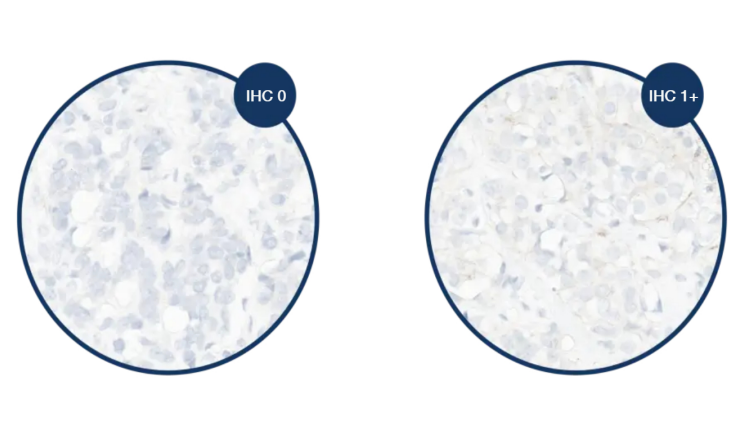
Identifying HER2-low expression relies on two important factors: the consistent distinguishing between IHC 0 and IHC 1+ and the seamless communication of discrete HER2 IHC scores between pathologists and oncologists.[2] Differentiating these staining patterns can be difficult; however, reliable reporting of scores at the low end of the spectrum may be clinically meaningful for patients with breast cancer.[1]

HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry.

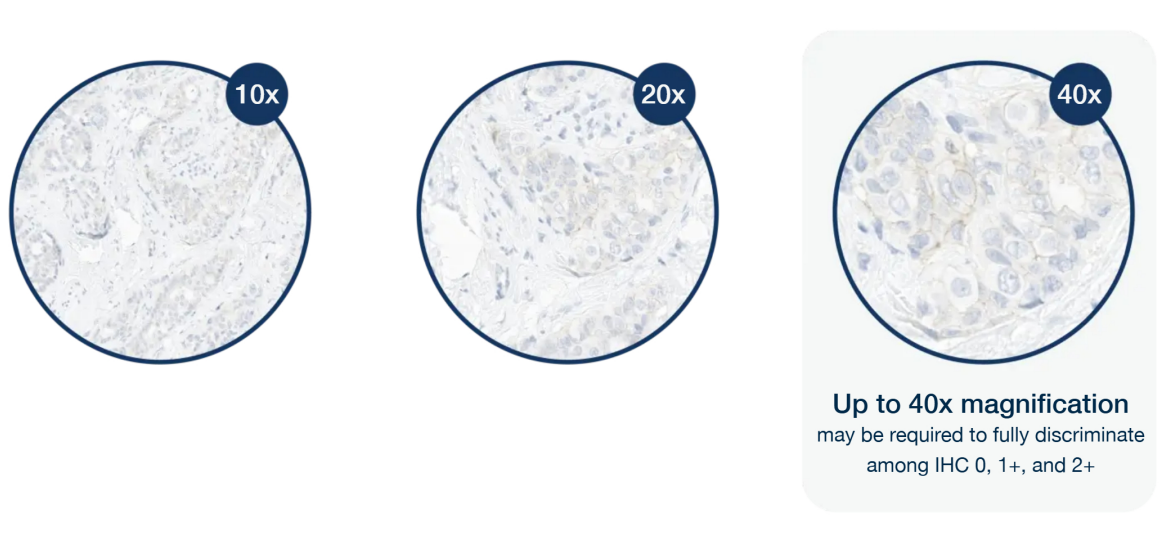
Interpret and Classify HER2 Expression Status Using ASCO/CAP Testing Guidelines[2]

The 2023 ASCO/CAP guideline update provides recommendations for interpreting and reporting low expression of HER2.[2] The ASCO/CAP guidelines define HER2 IHC 0 as either no staining is observed or membrane staining that is incomplete and is faint/barely perceptible and in ≤10% of tumor cells.[2,21] Meanwhile, IHC 1+ is defined as incomplete membrane staining that is faint/barely perceptible and in >10% of tumor cells.[2]

Samples on the border of IHC 0 and 1+ can be difficult to interpret



Cases can be examined at 40x or higher power magnification to discriminate between IHC 0 and 1+.[2]



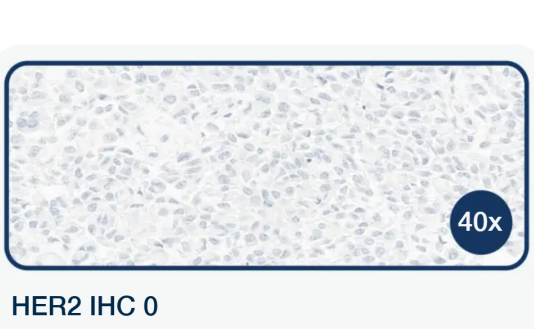
Up to 40x magnification may be required to fully discriminate among IHC 0, 1+, and 2+

Images contributed from HER2Know.com.[19]

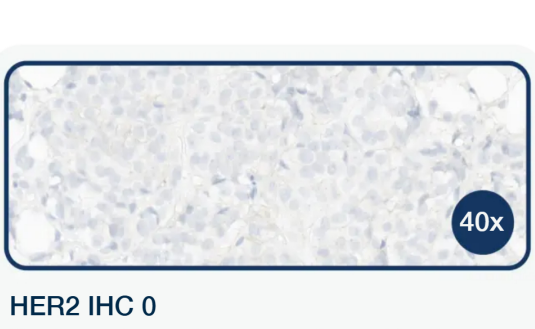
Tips for differentiating between IHC 0 and IHC 1+[2]:

- Examine HER2 IHC-stained slides using standardized ASCO/CAP guidelines scoring criteria
- Examine HER2 IHC at high power magnification (40x) when discriminating 0 from IHC 1+ staining
- Consider a second pathologist review when results are close to the 0 versus IHC 1+ interpretive threshold (>10% of cells with incomplete membrane staining that is faint/barely perceptible)
- Use controls with a range of protein expression (including 1+) to help ensure the assay has an appropriate limit of detection
- Give careful attention to preanalytical conditions of breast cancer tissue samples from both primary and metastatic sites

High power magnification (40x) may also help differentiate the distinct types of HER2 IHC 0 results (with and without staining):



HER2 IHC 0
No staining is observed



HER2 IHC 0
Membrane staining that is incomplete and faint/barely perceptible, and in ≤10% of tumor cells

Images contributed from HER2Know.com.[19]
ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry.

Suboptimal Preanalytic Factors Can Prevent Consistent Interpretation of HER2 Expression Levels in Breast Cancer[4]

Sample collection

- Core needle biopsies may not account for heterogeneity, and are prone to edge effects and crush artifacts[2,22]
- If the initial HER2 test result in a core needle biopsy is negative or equivocal, a new HER2 test may be ordered on the excision specimen[2]
- Scan the entire tissue section to ensure scoring is carried out on well-preserved and well-stained areas[2]

HER2 signal

- Time lapse decreases HER2 signal intensities[2,22]

Time to fixation

- Delays in time from tissue acquisition to fixation (i.e., cold ischemic time) can compromise validity of HER2 test results[2,22]

Fixation consistency

- Fixation variables may artificially drive down the detection of protein expression[2,22]
- Keep time to fixation as short as possible; specimens should be sliced into 5–10 mm intervals and fixed in 10% NBF for 6–72 hours[2]

Artifacts

- An awareness of potential artifacts is important to avoid over-interpretation of aberrant staining[22]

Quality assurance

- Validate HER2 IHC assays according to the most recent ASCO/CAP guidelines[2]
- Request additional tests if HER2 IHC cannot be determined due to lack of appropriate controls, evidence of inadequate specimen handing, artifacts, or analytic testing failure[2]

Find best practices to address sample limitations.

Browse Testing Solutions

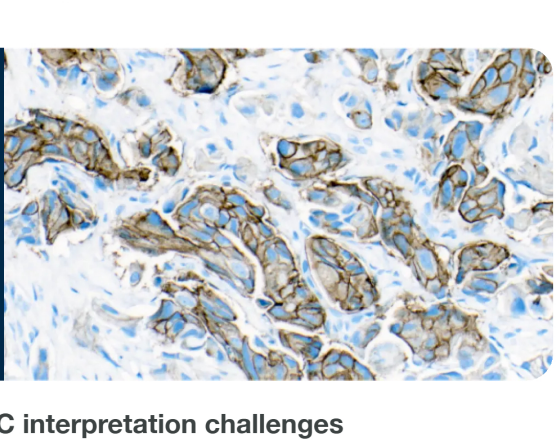
ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; NBF, neutral buffered formalin.

Guideline Recommendations Can Provide Consistency When Interpreting Challenging HER2 IHC Results[2]

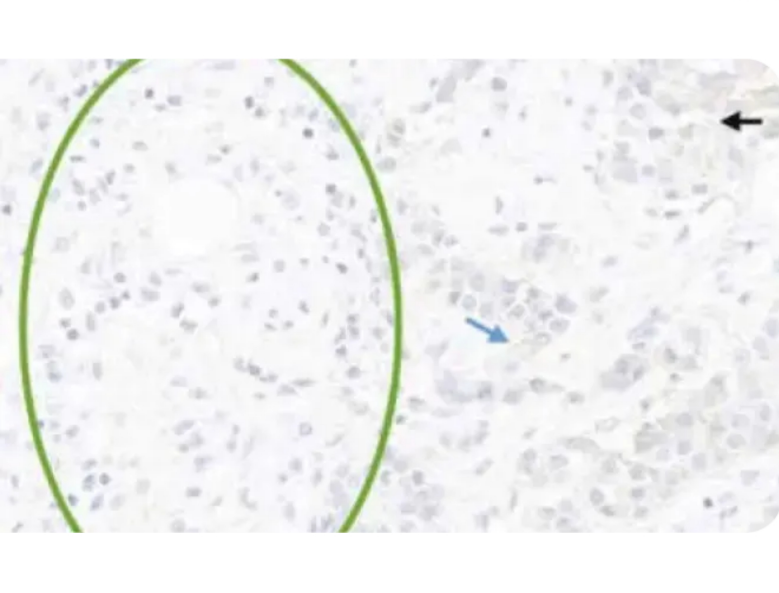
Adhering to 2023 ASCO/CAP guideline recommendations can help ensure consistency when interpreting HER2 expression results by IHC.[2]

Find out more about guideline recommendations for HER2 testing.

Browse Latest Developments



Solving common HER2 IHC interpretation challenges

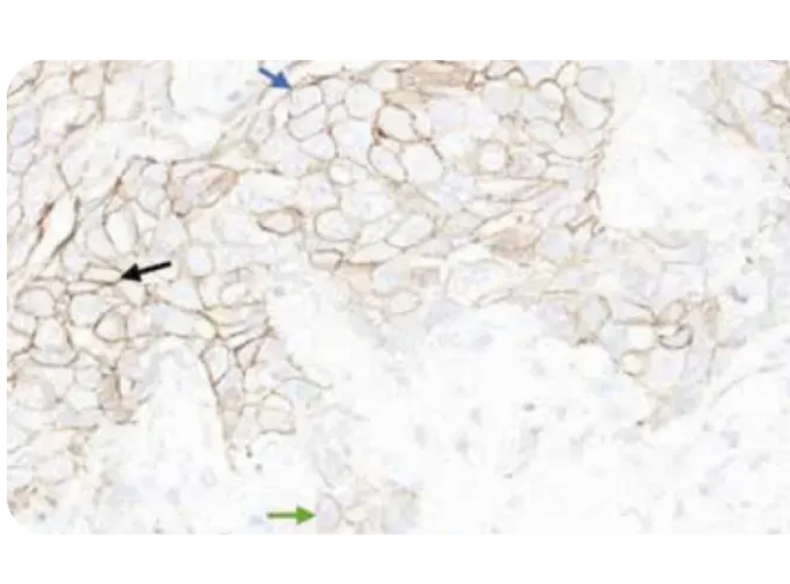


Distinguishing membrane staining from cytoplasmic, basal only, and nuclear staining can make HER2 IHC interpretation challenging.[23]

IHC 1+ (20x original magnification). About 15% of the tumor cells form a distinct focus displaying a faint and incomplete membranous staining (black arrow), while the rest of the tumor is negative (green circle). Some of the tumor cells display a non-specific, faint cytoplasmic blush (blue arrow) which does not affect evaluation.

Heterogeneous staining on tumor samples characterized by presence of cells with different levels of HER2 staining can make HER2 IHC interpretation challenging.[23,24]

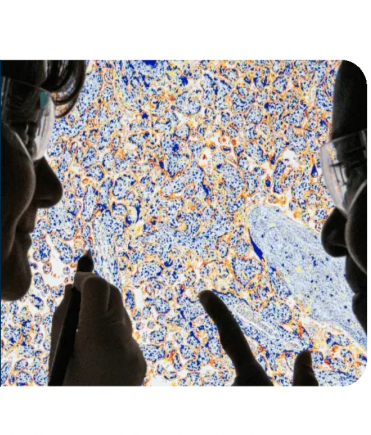
Heterogeneous staining, IHC 3+ spanning a range of patterns and intensities from strong complete (25% black arrow) to moderate complete (50% blue arrow) and weak incomplete (20% green arrow).



Images contributed from HER2Know.com.[19]

Access educational resources, clinical cases and tools to assess your HER2 IHC scoring consistency in solid tumors.

Visit HER2Know.com



ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry.

CAP Guidelines Help Ensure Standardized HER2 IHC Reporting[2]

Standardization of assay results relies on adherence to proficiency and accreditation requirements.[2] CAP recommends that testing laboratories perform ongoing competency assessment and proficiency tests at least twice a year (with performance requirements starting at 90%). Further recommendations include laboratory accreditation as well as onsite inspection every other year and self-inspection every year.[2]

How HER2 results are reported can have implications for patient care.[25] As many as 50% of pathology reports for breast cancer are missing some elements critical to patient management.[1]

To ensure reliable differentiation of HER2-low expression, record discrete scores on each patient's HER2 IHC report. Recording staining patterns, such as faint/barely perceptible/incomplete staining, and percentage staining is also important.[11]

Recommended result reporting

IHC 0 HER2-negative	IHC 1+ HER2-negative[a]	IHC 2+/ISH- HER2-negative[a]	IHC 2+/ISH+ HER2-positive	IHC 3+ HER2-positive
	ASCO/CAP breast biomarker reporting template guidance[11] *[a]Breast cancers with HER2 IHC score 1+ or HER2 IHC score 2+ and a negative ISH result may be reported as 'HER2-low'*			

Cannot be determined or test not performed[2]

Must report HER2 test result as indeterminate if technical issues prevent one or both tests (IHC and ISH) from being reported as positive, negative or equivocal.
Conditions may include:

- Inadequate specimen handling
- Artifacts (crush or edge artifacts) that make interpretation difficult
- Analytic testing failure

Another specimen should be requested for testing to determine HER2 status. Reason for indeterminate testing should be noted in a comment in the report.

ASCO/CAP comment recommendation[2]

Please refer to 'The Bottom Line' in the 2023 ASCO/CAP guidelines update for HER2 testing report footnote comment recommendation.

CAP has developed pathology reporting templates to promote complete and standardized reporting of breast cancer specimens for HER2.[11] The National Comprehensive Cancer Network® (NCCN®) Breast Cancer Panel endorses the use of CAP protocols for reporting the pathologic analysis of all breast cancer specimens.[1]

[a]Breast cancers with HER2 IHC score 1+ or HER2 IHC score 2+ and a negative ISH result are eligible for clinically appropriate HER2-targeted therapy and may be reported as HER2-low.
ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization; NCCN, National Comprehensive Cancer Network.

Find out how to improve HER2 IHC scoring to ensure reliable differentiation of low levels of HER2 expression

Learn more about identifying HER2-low in breast cancer and assigning a HER2 IHC score when encountering challenging staining patterns.

Visit HER2Know.com

HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry.

Considerations for Your Practice

HER2 testing has become routine practice to ensure all patients with breast cancer receive more informed treatment options. Reliable differentiation of scores at the low end of the spectrum may be clinically meaningful for patients with breast cancer.[1,2]

- Follow ASCO/CAP best practices for HER2 testing; examine HER2 IHC at high power magnification (40x) when discriminating IHC 0 from 1+ staining; IHC with reflex ISH for 2+ specimens is the recommended methodology[2]
- Score consistently across the full spectrum of HER2 expression; in lower levels of HER2 expression, differentiating between HER2 IHC 0 and 1+ supports active research and refinement of testing criteria[2]
- Use the CAP reporting template to ensure HER2 test results are in an ideal format for interpretation by the MDT[11]

ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry; ISH, *in situ* hybridization; MDT, multidisciplinary team.

Helpful Resources

Check out these featured resources on HER2 biomarker testing in breast cancer.

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FOR HCPS
Identifying low levels of HER2 expression in breast cancer
Find out about College of American Pathologists' (CAP) 2023 updated reporting template for human epidermal growth factor 2 (HER2)-low breast cancer and best practices for pathologists when reviewing these cases.
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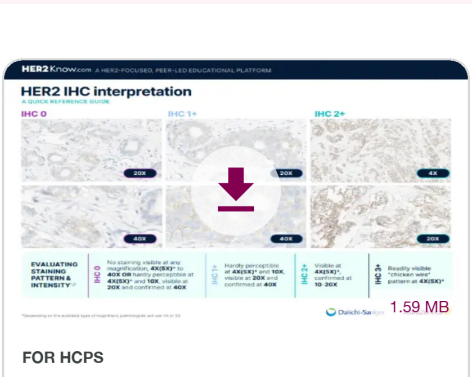

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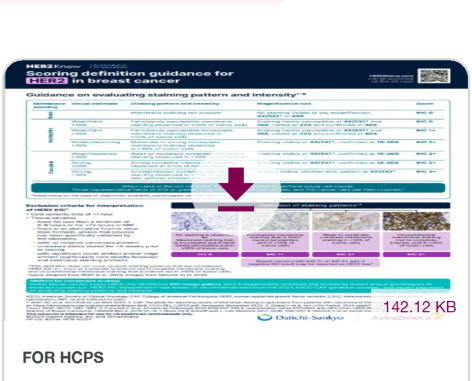

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Download this resource for a summarized approach to human epidermal growth factor 2 (HER2) immunohistochemistry (IHC) scoring interpretation based on the 2018 American Society of Clinical Oncology (ASCO)-
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ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; IHC, immunohistochemistry.

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HER2, human epidermal growth factor receptor 2.

References

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