Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 1

Shi Wei,
MD, PhD

Professor of Pathology and Laboratory Medicine, The Barbara F. Atkinson Endowed Professor, Director of Translational Research,
University of Kansas Medical Center

This is a synopsis of session 1 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

From past to present: The significance of HER2 in Breast Cancer

Key takeaways

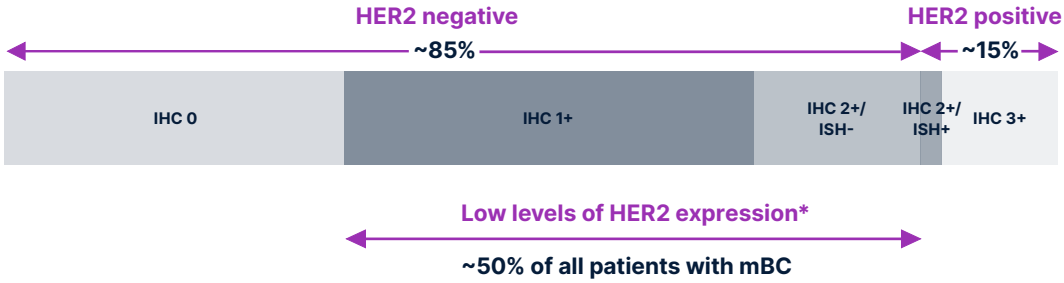
- **HER2 is a prognostic and predictive biomarker for breast cancer**[1]

  The binary classification of HER2 status has historically informed use of HER2-targeted therapies, but may not account for the full spectrum of HER2 expression

- **The spectrum of HER2 expression allows classification beyond positive and negative**

  HER2 expression exists on a spectrum and while 85% of breast cancers are currently identified as HER2 negative, approximately 50% of all patients with breast cancer may exhibit low levels of HER2 expression[1]

  - Most of the published data and ongoing clinical trials define low levels of HER2 expression as IHC 1+, IHC 2+/ISH-[1]



*Low levels of HER2 expression are not currently classified within the 2018 ASCO/CAP guidelines, but are recognized as HER2 negative.[1]

- **HER2 expression is not static, but dynamic over the course of disease**

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

1. Tarantino P, et al. J Clin Oncol. 2020;38(17):1951–62.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 1

Savitri Krishnamurthy, MD

Professor of Pathology,
The University of Texas MD Anderson Cancer

Hannah Wen, MD, PhD

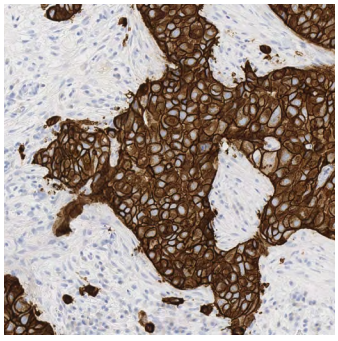
Director, Breast Pathology Fellowship,
Memorial Sloan Kettering Cancer Center

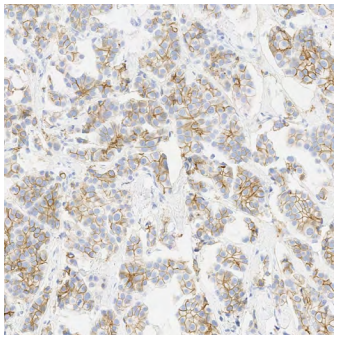
This is a synopsis of session 1 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

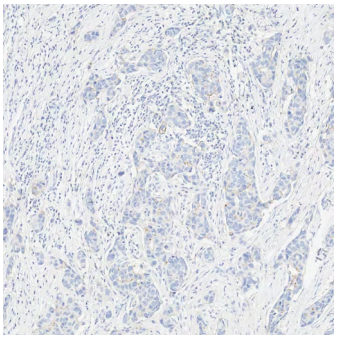
Microscopy case walkthrough HER2 IHC: From common to unusual staining patterns

Key takeaways[1]

- Always review the corresponding H&E slide together with the IHC to ensure correlation of tumor morphology and staining pattern
- Recognize unique staining patterns, such as the basolateral staining pattern of invasive micropapillay carcinoma
- HER2-expressing tumors that are classified as IHC negative include a range of staining; from no membrane staining or faint, incomplete staining in ≤10% of tumor cells (IHC 0), to faint, incomplete staining in >10% of tumor cells (IHC 1+)







To view additional clinical cases, please visit HER2Know.com

HER2, human epidermal growth factor receptor 2; H&E, hematoxylin and eosin stain; IHC, immunohistochemistry.

1. AstraZeneca and Daiichi Sankyo. Data on file. REF-20181. 2022.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services.

The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.

PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 2

David Hicks, MD

Professor and Director of IHC-ISH Laboratory and Breast Subspecialty Service,
University of Rochester Medical Center

This is a synopsis of session 2 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Consistency with practice: Standardizing the approach to HER2 IHC scoring

Key takeaways

Standardization of all components of HER2 testing is critical to help ensure consistent, reliable, and clinically actionable results. Variability in clinical interpretation of HER2 determination can be minimized through standardization of testing processes and approaches to scoring and evaluation of HER2 expression.[1]

2022 updates to the CAP accreditation program checklists capture significant changes to further improve pre-analytic quality of specimens.[2]

In the **post-analytic setting,** ASCO/CAP interpretation criteria, validated image analysis, reporting elements, and quality assurance procedures are significant considerations in the interpretation of HER2 test results.[3,4]

Maintaining good practice[3,5,6]

- Establish best practice of second pathologist read for borderline or challenging cases with IHC 0 or 1+ or cases with intratumoral heterogeneity
- Ongoing monitoring of scoring reproducibility and concordance
- Utilize minimum required tumor cell count (i.e., minimum of 100 cells), and if not present, repeat test on a subsequent specimen
- Routine use of cell line control samples to improve quality HER2 differentiation
- Conduct trend analysis. Track overall HER2 IHC 1+ rates (100 random cases and re-scoring to assess rates of IHC 0 and 1+)

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

1. Hicks DG, et al. Lab Medicine 2011;42:459–467. 2. CAP TODAY Sep 2021 Issues: A preanalytics push in accreditation checklists. 3. Wolff AC, et al. J Clin Oncol 2018;36:2105–2122. 4. CAP template for reporting results of biomarker testing of specimens from patients with carcinoma of the breast. 5. AstraZeneca and Daiichi Sankyo. Data on file. REF-8201. 2021. 6. AstraZeneca and Daiichi Sankyo. Data on file. REF-20250. 2022.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 2

Gary Tozbikian, MD

Associate Professor and Breast Pathology Division Director,
The Ohio State University Wexner Medical Center

Thaer Khoury, MD

Vice Chair of Anatomic Pathology, Chief of Breast Pathology,
Roswell Park Comprehensive Cancer Center

This is a synopsis of session 2 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Microscopy case walkthrough HER2 IHC: Artifacts and indeterminate staining patterns

Key takeaways[1]

Crush artifact

- Distorted tissue with elongation and hyperchromasia of epithelial cells nuclei, tissue fragmentation, and/or loss of membranous, nuclear and cytoplasmic details
- During evaluation, avoid the affected area; if all tumor cells are affected by crush artifact, request another biopsy

Ink artifact

- Occurs when ink is applied to surgical margins during specimen processing; presence of ink may obscure HER2 IHC interpretation
- During evaluation avoid the inked margins and evaluate more preserved, un-inked tissue for IHC staining

Edge artifact

- Accentuation of non-specific staining at periphery of tissue, occurs when there is inconsistent fixation, tissue drying or lifting around edges
- During evaluation avoid interpretation in areas of the edge artifact, or make necessary changes to fixation time and/or storage time or environment of cut slides[2]

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 2

Gary Tozbikian, MD

Associate Professor and Breast Pathology Division Director,
The Ohio State University Wexner Medical Center

Thaer Khoury, MD

Vice Chair of Anatomic Pathology, Chief of Breast Pathology,
Roswell Park Comprehensive Cancer Center

This is a synopsis of session 2 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Other artifacts and indeterminate staining patterns

- Uneven antibody dispensing
- Antibody concentrating at edge of slide
- Prolonged cold ischemia time
- Poor quality external positive controls
- Cytoplasmic staining
- Decalcification effects

It is advisable to seek a second expert opinion when specimen staining issues, including, antibody concentration and dispersal, edge artifacts, prolonged ischemia time, or decalcification are suspected. Furthermore, adherence to CAP documentation guidelines, and proactive and effective communication with the multidisciplinary breast cancer care team are strongly recommended.

After exhausting all options available on the tumor tissue with artifacts and indeterminate staining patterns, requesting an additional biopsy could be essential to reach proper HER2 classification.

To view additional clinical cases, please visit HER2Know.com

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

1. AstraZeneca and Daiichi Sankyo. Data on file. REF-20251. 2022. 2. Haragannavar VC et al. World Journal of Dentistry 2018;9(4):333-341.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 3

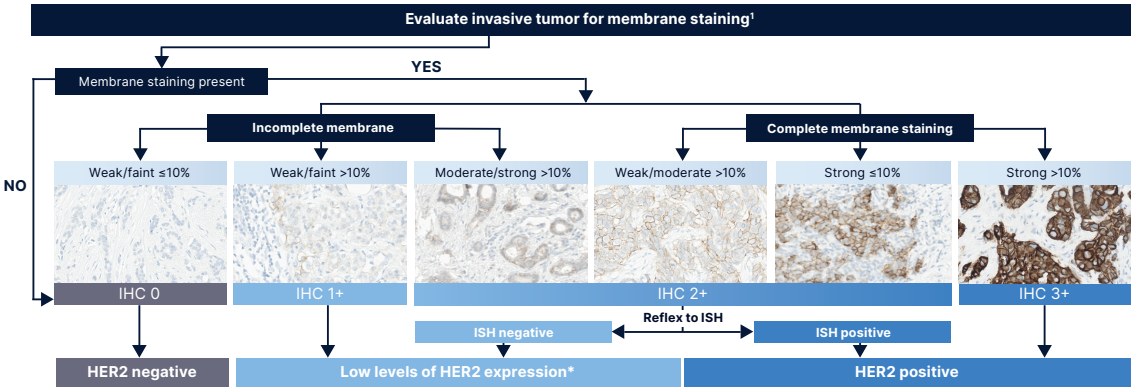
Shabnam Jaffer, MD

Professor of Pathology, Molecular and Cell Based Medicine,
The Icahn School of Medicine at Mount Sinai

This is a synopsis of session 3 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Challenge accepted: Best practices for scoring HER2 IHC across the spectrum

Key takeaways



***Low levels of HER2 expression are not currently classified within the 2018 ASCO/CAP guidelines, but are recognized as HER2 negative.[2]**

Tumor heterogeneity is an important feature of breast cancer and may impact concordance and reproducibility.[3] Intratumoral heterogeneity may be observed in 6-36% of advanced tumors and is significantly more common in cases with HER2 equivocal status by ISH and/or IHC.[4–10]

Likely features with corresponding HER2 expression

HER2 negative	Low levels of HER2 expression* (IHC 1+ or IHC 2+/ISH-)[2]
• Usually luminal A[11] • Low grade indolent triple negative cancers such as adenoid cystic carcinoma, acinic cell carcinoma, secretory carcinoma, fibromatosis-like metaplastic carcinoma, low-grade adenosquamous carcinoma, mucoepidermoid carcinoma[12]	• Heterogeneous group[13] • More frequently found within HR-positive disease (65.4%) compared to TNBC (36.5%)[13] • Older patients, larger tumor size, more nodal involvement[13]

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 3

Shabnam Jaffer, MD

Professor of Pathology, Molecular and Cell Based Medicine,
The Icahn School of Medicine at Mount Sinai

This is a synopsis of session 3 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Staining pattern, intensity, and visual estimations should be scrutinized at appropriate magnifications to identify low levels of HER2 expression.[14]

	Recommended magnification	Score
Weak/faint ≤10%	Staining hardly perceptible at 4X(5X)* and 10X, visible at 20X and confirmed at 40X	IHC 0
Weak/faint >10%	Staining hardly perceptible at 4X(5X)* and 10X, visible at 20X and confirmed at 40X	IHC 1+
Moderate/strong >10%	Staining visible at 4X(5X)*, confirmed at 10-20X	IHC 2+
Weak/moderate >10%	Staining visible at 4X(5X)*, confirmed at 10-20X	IHC 2+
Strong ≤10%	Staining visible at 4X(5X)*, confirmed at 10-20X	IHC 2+
Strong >10%	Staining visible at 4X(5X)*	IHC 3+

When close to the cut off point, count 100 cells in three representative fields, to establish percentage, at 40X or higher.[15]

ASCO, American Society of Clinical Oncology; CAP, College of American Pathologists; HER2, human epidermal growth factor receptor 2; HR, hormone receptor; IHC, immunohistochemistry; ISH, in situ hybridization; TNBC, triple negative breast cancer.

1. AstraZeneca and Daiichi Sankyo. Data on file. REF-18013. 2021. 2. Wolff AC, et al. J Clin Oncol 2018; 36(20):2105–2121. 3. Marchiò C, et al. Semin Cancer Biol 2020;72:123–35. 4. Allison K, et al. Am J Clin Pathol 2011;136:864–71. 5. Bartlett A, et al. Am J Clin Pathol 2011;136:266–74. 6. Chang M, et al. Modern Pathology 2012;25:683–88. 7. Lee H, et al. Am J Clin Pathol 2014;142:755–66. 8. Lee H, et al. Am J Clin Pathol 2015;144:570–8. 9. Seol H, et al. Modern Pathology 2012;25:938–48. 10. Bethune G, et al. Ann Diagn Pathol 2015;19:385–90. 11. American Cancer Society, Inc., Surveillance Research Breast Cancer Facts & Figures 2019-2020. 12. Cserni G, et al. Cancers 2021;13(22):5964. 13. Schettini F, et al. NPJ Breast Cancer 2021;7:1. 14. Franchet C, et al. Annales de Pathologie 2021;41(6):507–20. 15. Interpretation Guide PATHWAY anti-HER-2/neu (4B5) Rabbit Monoclonal Primary Antibody Staining of Breast Carcinoma, 1499100 Rev K, 20.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services.

The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.

PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca

Interpreting HER2 in Breast Cancer

A full spectrum of possibilities

Short Course
Session 3

Savitri Krishnamurthy, MD

Professor of Pathology,
The University of Texas MD Anderson Cancer Center

Marilyn Bui, MD, PhD

Senior Member & Professor in the Department of Pathology,
Moffitt Cancer Center & Research Institute

This is a synopsis of session 3 of the short course, Interpreting HER2 in Breast Cancer: A full spectrum of possibilities. Please visit the USCAP website to view the full 3-part course in its entirety.

Microscopy case walkthrough HER2 IHC: Challenging/borderline cases

Key takeaways[1]

Borderline HER2 expression (between IHC 0 vs 1+)

- This distinction can be challenging and subjective, with suggested careful evaluation at both medium and high-power objectives. Consensus review is a valuable avenue of assessment of these cases

Borderline HER2 expression (between IHC 1+ and 2+)

- Incomplete membrane staining (weak to moderate intensity in >10% of tumor cells) can be mistaken for HER2 IHC 2+ if the complete membranous staining pattern is focal or when complete staining is not recognized; ancillary HER2 ISH testing may be performed in such borderline cases

Core biopsy vs subsequent surgical resection

- If the initial HER2 test result in a core needle biopsy specimen of a primary breast cancer is negative, a new HER2 test may be ordered on the excision specimen if one of the following is observed[2]:
  - Tumor is grade 3
  - Amount of invasive tumor in the core biopsy specimen is small
  - Resection specimen contains high-grade carcinoma that is morphologically distinct from that in the core
  - Core biopsy result is equivocal for HER2 after testing by both ISH and IHC
  - There is doubt about the handling of the core biopsy specimen (long ischemic time, short time in fixative, different fixative) or the test is suspected by the pathologist to be negative on the basis of testing error

To view additional clinical cases, please visit HER2Know.com

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

1. AstraZeneca and Daiichi Sankyo. Data on file. REF-20318. 2022. 2. Wolff AC, et al. J Clin Oncol 2018; 36(20):2105–2121.

The information presented represents the opinion of the presenters and is not necessarily the view of USCAP. USCAP does not endorse the products or services. The short course is possible through a sponsorship from AstraZeneca and Daiichi Sankyo.

This resource is intended for use by US healthcare professionals only.
©2022 Daiichi Sankyo, Inc, and AstraZeneca.
PP-US-8201a-1448 08/22

Daiichi-Sankyo | AstraZeneca