ASCO Guidelines

COLLEGE of AMERICAN PATHOLOGISTS

Human Epidermal Growth Factor Receptor 2 (HER2) Testing in Breast Cancer Guideline: 2018 Focused Update

Summary of Changes and Definitions

Recommendations previously published in response to letter to editor[1]

1. The revised definition of IHC 2+ (equivocal) is invasive breast cancer with "weak to moderate complete membrane staining observed in >10% of tumor cells." (Revised Figure 1)

2. 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: (Revised Table 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

The evidence quality is high and the strength of recommendation is strong.

New recommendations with ISH Group Definitions HER2/CEP17 ratio to HER2 signals/cell

Group 1	Group 2	Group 3	Group 4	Group 5
Ratio ≥2.0 ≥4.0 signals/cell	Ratio ≥2.0 <4.0 signals/cell	Ratio <2.0 ≥6.0 signals/cell	Ratio <2.0 ≥4.0 and <6.0 signals/cell	Ratio <2.0 <4.0 signals/cell

3. (Group 2) If a case has a *HER2*/CEP17 ratio of ≥ 2.0 but the average *HER2* signals/cell is < 4.0, a definitive diagnosis will be rendered based on additional work-up. If not already assessed by the institution or laboratory performing the ISH test, IHC testing for HER2 should be performed using sections from the same tissue sample used for ISH, and the slides from both ISH and IHC should be reviewed together to guide the selection of areas to score by ISH (local practice considerations will dictate the best procedure to accomplish this concomitant assessment): (Figure 4)
   - **a.** If the IHC result is 3+, diagnosis is HER2 positive
   - **b.** If the IHC result is 2+, recount ISH by having an additional observer, blinded to previous ISH results, count at least 20 cells that include the area of invasive cancer with IHC 2+ staining:
     - If reviewing the count by the additional observer changes the result into another ISH category, the result should be adjudicated per internal procedures to define the final category.
     - If the count remains an average of < 4.0 HER2 signals/cell and the HER2/CEP17 ratio is ≥ 2.0, diagnosis is HER2 negative with a comment.*****
   - **c.** If the IHC result is 0 or 1+, diagnosis is HER2 negative with a comment.*****

     ***Comment:** Evidence is limited on the efficacy of HER2-targeted therapy in the small subset of cases with a HER2/CEP17 ratio of ≥2.0 and an average HER2 copy number of < 4.0 per cell. In the first generation of adjuvant trastuzumab trials, patients in this subgroup who were randomly assigned to the trastuzumab arm did not seem to derive an improvement in disease-free or overall survival, but there were too few such cases to draw definitive conclusions. IHC expression for HER2 should be used to complement ISH and define HER2 status. If the IHC result is not 3+ positive, it is recommended that the specimen be considered HER2 negative because of the low HER2 copy number by ISH and the lack of protein overexpression.

4. (Group 3) If a case has an average of ≥ 6.0 HER2 signals/cell with a HER2/CEP17 ratio of < 2.0, formerly diagnosed as ISH positive for HER2, a definitive diagnosis will be rendered based on additional work-up. If not already assessed by the institution or laboratory performing the ISH test, IHC testing for HER2 should be performed using sections from the same tissue sample used for ISH, and the slides from both ISH and IHC should be reviewed together to guide the selection of areas to score by ISH (local practice considerations will dictate the best procedure to accomplish this concomitant review): (Figure 5)

a. If the IHC result is 3+, diagnosis is HER2 positive

b. If the IHC result is 2+, recount ISH by having an additional observer, blinded to previous ISH results, count at least 20 cells that include the area of invasion with IHC 2+ staining:

- If reviewing the count by the additional observer changes the result into another ISH category, the result should be adjudicated per internal procedures to define the final category
- If the *HER2*/CEP17 ratio remains < 2.0 with ≥ 6.0 *HER2* signals/cell, diagnosis is HER2 positive

c. If the IHC result is 0 or 1+, diagnosis is HER2 negative with a comment*

***Comment:** There are insufficient data on the efficacy of HER2-targeted therapy in cases with a HER2 ratio of < 2.0 in the absence of protein overexpression because such patients were not eligible for the first generation of adjuvant trastuzumab clinical trials. When concurrent IHC results are negative (0 or 1+), it is recommended that the specimen be considered HER2 negative.

5. (Group 4) If the case has an average HER2 signals/tumor cell of ≥ 4.0 and < 6.0 and the HER2/CEP17 ratio is < 2.0, formerly diagnosed as ISH equivocal for HER2, a definitive diagnosis will be rendered based on additional work-up. If not already assessed by the institution or laboratory performing the ISH test, IHC testing for HER2 should be performed using sections from the same tissue sample used for ISH, and the slides from both ISH and IHC should be reviewed together to guide the selection of areas to score by ISH (local practice considerations will dictate the best procedure to accomplish this concomitant review): (Figure 6)

a. If the IHC result is 3+, diagnosis is HER2 positive

b. If the IHC result is 2+, recount ISH by having an additional observer, blinded to previous ISH results, count at least 20 cells that include the area of invasion with IHC 2+ staining:

- If reviewing the count by the additional observer changes the result into another ISH category, the result should be adjudicated per internal procedures to define the final category
- If the count remains an average of ≥ 4.0 and < 6.0 HER2 signals/cell with a *HER2*/CEP17 ratio of < 2.0, diagnosis is HER2 negative with a comment*****

c. If the IHC result is 0 or 1+, diagnosis is HER2 negative with a comment*****

***Comment:** It is uncertain whether patients with an average of ≥ 4.0 and < 6.0 HER2 signals per cell and a HER2/CEP17 ratio of , 2.0 benefit from HER2 targeted therapy in the absence of protein overexpression (IHC 3+). If the specimen test result is close to the ISH ratio threshold for positive, there is a high likelihood that repeat testing will result in different results by chance alone. Therefore, when IHC results are not 3+ positive, it is recommended that the sample be considered HER2 negative without additional testing on the same specimen.

For all ISH group recommendations the evidence quality is intermediate and the strength of recommendation is strong.

Summary of HER2 ISH Diagnostic Criteria

HER2 Positive	HER2 Negative
Dual Probe Assay	
Group 1	Group 2 AND concurrent IHC 0-1+ or 2+
Group 2 AND concurrent IHC 3+	Group 3 AND concurrent IHC 0-1+
Group 3 AND concurrent IHC 2+ or 3+	Group 4 AND concurrent IHC 0-1+ or 2+
Group 4 AND concurrent IHC 3+	Group 5
Single Probe Assay	
HER2 copy number ≥6.0 signals/cell	HER2 copy number <4.0 signals/cell
HER2 copy number ≥4.0 and <6.0 signals/cell AND concurrent IHC 3+	HER2 copy number ≥4.0 and <6.0 signals/cell AND concurrent IHC 0 or 1+
HER2 copy number ≥4.0 and <6.0 signals/cell AND concurrent dual probe Group 1	HER2 copy number ≥4.0 and <6.0 signals/cell AND concurrent dual probe Group 5

Group 1	Group 2	Group 3	Group 4	Group 5
Ratio ≥2.0 ≥4.0 signals/cell	Ratio ≥2.0 <4.0 signals/cell	Ratio <2.0 ≥6.0 signals/cell	Ratio <2.0 ≥4.0 and <6.0 signals/cell	Ratio <2.0 <4.0 signals/cell

1. Wolff AC, Hammond ME, Hicks DG, et al: Reply to E.A. Rakha et al. *J Clin Oncol* 33:1302-4, 2015

Wolff AC, Hammond EH, Allison KH, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/ College of American Pathologists Clinical Practice Guideline Focused Update. *Arch Pathol Lab Med*. 2018;142(11):1364-1382.