ERBB2 (*HER2/neu*) Gene Amplification by FISH with Reflex, Tissue

Indications for Ordering

- Aid in prediction of response to HER2-directed therapy [eg, trastuzumab (Herceptin)] in patients with breast or gastric cancer
- Confirm equivocal HercepTest (2+) immunohistochemistry (IHC) result

Test Description

- Two-color fluorescence in situ hybridization (FISH) for the determination of ERBB2 (HER2) copy number
- FDA approved for invasive breast cancer and gastric cancer patients who are potential candidates for trastuzumab therapy

Tests to Consider

Typical Testing Strategy

Standard practice for evaluating primary, recurrent, and metastatic breast carcinomas, and gastric or gastroesophageal carcinomas

- Assess *ERBB2* status by IHC or FISH
  - Concordance between the methods can vary due to subjective interpretation and method-specific limitations
- Use alternate test if equivocal results are reported on initial test
  - If IHC equivocal (2+), confirm by FISH
  - If FISH equivocal on breast specimen, confirm by IHC with rescoring in area(s) of interest if IHC is equivocal (2+)
  - If FISH equivocal on gastric specimen, confirm result with an alternate control probe or an alternative analytic method

Primary Test

ERBB2 (*HER2/neu*) Gene Amplification by FISH with Reflex, Tissue 2008603

- Use to confirm equivocal HercepTest IHC result (2+)

Related tests

Measurement of protein expression

ERBB2 (HER2/neu) (HercepTest) with Interpretation by Immunohistochemistry, Tissue 0049174

ERBB2 (HER2/neu) (HercepTest) by Immunohistochemistry, Tissue with Reflex to FISH if 2+ 0049178

ERBB2 (HER2) (HercepTest) by Immunohistochemistry 2007332

Disease Overview

Incidence

Approximately 234,000 new invasive breast and 24,500 new gastric cancers (NCCN, 2015) are diagnosed in the U.S. per year; common causes of cancer-related deaths

Treatment issues

- Amplification of the *ERBB2* gene occurs in 15-20% of breast cancers and approximately 20% of gastric cancers
  - Predicts poor prognosis in invasive breast cancer
  - Trastuzumab prolongs the overall survival rate in individuals with breast or gastric cancer when tumors overexpress HER2
  - Trastuzumab antibodies are directed against the extracellular portion of the HER2 protein
    - Inhibits HER2-overexpressing cancers
- Due to high drug costs and cardiac toxicity, use of trastuzumab requires identification of tumors that demonstrate *ERBB2 (HER2)* gene amplification or protein overexpression (3+ IHC result)
- New therapies targeting HER2 include pertuzumab (Perjeta), T-DM1 (Kadcyla), and lapatinib (Tykerb)
- A recent study showed that treatment with a combination of trastuzumab and pertuzumab was more effective than trastuzumab alone (in combination with docetaxel) in prolonging survival of breast cancer patients

Genetics

Gene: *ERBB2*

Function: amplification of *ERBB2* gene

- Increases membrane expression and activation of the HER2 protein
- Stimulates cell proliferation

Test Interpretation

Results

Breast

- Positive: *ERBB2*/CEP17 ratio >2.0 and average ERBB2 copy number >4.0 signals/cell (Group 1)
  - Predicts favorable response to targeted therapy
- Negative: *ERBB2*/CEP17 ratio <2.0 and average ERBB2 copy number <4.0 signals/cell (Group 5)
  - Predicts lack of response to targeted therapy
- Indeterminate: *ERBB2*/CEP17 ratio >2.0, and average *ERBB2* copy number <4.0 (Group 2); *ERBB2*/CEP17 ratio <2.0, and average *ERBB2* copy number ≥6.0 signals/cell (Group 3); *ERBB2*/CEP17 ratio <2.0, and average *ERBB2* copy number ≥4.0 and <6.0 signals/cell (Group 4)
  - For groups 2-4 by *ERBB2*/CEP17, concomitant Her2 IHC review is performed
  - An IHC score of 3+ is considered positive and 0 or 1+ is considered negative
  - For an IHC score of 2+, additional tumor nuclei are enumerated with FISH from area of highest IHC intensity by an individual blinded to the original results
  - Repeat scoring consistent with groups 2 and 4 is considered negative while scoring consistent with group 3 is considered positive
- It is uncertain whether patients with ≥4.0 and <6.0 average HER2 signals/cell and *HER2*/CEP17 ratio <2.0 benefit from HER2 targeted therapy in the absence of protein overexpression (IHC 3+)

Gastric

  - If equivocal by both *ERBB2*/CEP17 and HercepTest (IHC), confirm equivocal cases with an alternate control probe
  - If equivocal result has been obtained on a gastric biopsy, follow up testing should be performed on the resection specimen

Limitations

- Testing using tissue fixed in alcohol-based or nonformalin fixatives has not been validated using this method
- Specimens placed in decal may have a false-negative result
- This assay has been validated and is FDA approved for invasive breast carcinoma and gastric cancers only
- *ERBB2* (*HER2/neu*) FISH has been validated as a laboratory developed test for other cancer types
- Guidelines for interpretation of HER2 status in nonbreast and gastric cancers has not been published
- Testing is interpreted according to ASCO/CAP 2018 Updated Guidelines for breast cancer and ASCO/CAP 2017 Guidelines for gastric cancer
- Repeat testing is recommended for discordant results

References

- Bartley AN, Washington MK, Ventura CB, Ismaila N, Colasacco C, Benson AB 3rd, Carrato A, Gulley ML, Jain D, Kakar S, Mackay HJ, Streutker C, Tang L, Troxell M, Ajani JA. HER2 Testing and Clinical Decision Making in Gastroesophageal Adenocarcinoma: Guideline From the College of American Pathologists, American Society for Clinical Pathology, and American Society of Clinical Oncology. Arch Pathol Lab Med. 2016 Dec;140(12):1345-1363 PudMed
- Wolff AC, Hammond MEH, Allison KH, Harvey BE, Mangu PB, Bartlett JMS, Bilous M, Ellis IO, Fitzgibbons P, Hanna W, Jenkins RB, Press MF, Spears PA, Vance GH, Viale G, McShane LM, Dowsett M. 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 May 30 PubMed