

HER2 Determination in Gastric or Gastroesophageal Carcinoma

Background Information

Gastric cancer is the second most common cause of cancer-related deaths worldwide.¹ In the United States, the American Cancer Society estimates that 21,000 cases of gastric cancer were diagnosed in 2010 with 10,570 patients dying from the disease (6,350 men and 4,220 women).¹ The five-year survival for advanced or metastatic gastric cancer is around 5-20%, while the median overall survival is less than 1 year.^{1,3} This dismal prognosis underscores the need for new treatments.

Recent studies have evaluated the role of human epidermal growth factor receptor 2 (HER2) in gastric cancer. This protein is a member of a family of receptors that have been shown to drive cell proliferation, adhesion, migration and differentiation.⁴ HER2 in breast cancer is known to be a marker of poor prognosis but is also predictive of response to the humanized monoclonal antibody trastuzumab (Herceptin®). Trastuzumab targets HER2 and induces antibody-dependent cellular cytotoxicity that inhibits HER2-mediated signaling, and prevents cleavage of the extracellular domain of HER2.⁵ Inclusion of this drug in combination with chemotherapy is now the standard of care in both early and metastatic breast carcinoma.⁶⁻⁸ There is growing support that HER2 is also a driver of tumorigenesis in gastric cancer, with between 7-34% of gastric carcinomas showing *HER2* amplification or overexpression.^{4,9,10} Although the literature is somewhat varied, at least some studies have suggested that *HER2* amplification/overexpression in gastric cancer is associated with poor outcomes and more aggressive disease.^{4,10}

Recently, the results of the Trastuzumab for Gastric Cancer (ToGA) study were published wherein the clinical efficacy and safety of trastuzumab in addition to chemotherapy as the first-line treatment in patient's with advanced gastric or gastroesophageal carcinoma were published. This study was

a phase 3, randomized controlled trial that involved patients from 24 countries with either gastric or gastroesophageal junction cancers that were shown to overexpress HER2 by immunohistochemistry (IHC) or be *HER2* amplified by fluorescence *in situ* hybridization (FISH). To be eligible, carcinomas were 3+ by IHC or FISH positive with a HER2: CEP17 ratio ≥ 2 . Participants were randomly assigned in a 1:1 ratio to receive either a chemotherapy regimen or chemotherapy in combination with intravenous trastuzumab; the primary endpoint was overall survival. The results showed a modest but statistically significant improvement in overall survival in those patients that received trastuzumab. The median overall survival was 13.8 months in those assigned to trastuzumab plus chemotherapy in comparison to 11.1 months in those who received chemotherapy alone ($p=0.0046$).

Clinical Indications

HER2 testing is offered as a molecular adjunctive test in the management of patients with either metastatic gastric or gastroesophageal invasive carcinoma for whom therapy with the humanized monoclonal antibody trastuzumab is considered. Immunohistochemistry is the primary methodology for testing and can be performed on either biopsies or resection tissues that are formalin-fixed and paraffin-embedded. If an equivocal result is obtained with IHC (see *Interpretation section below*), cases are reflexed for *HER2* FISH testing, which is also performed on formalin-fixed and paraffin-embedded tissue. IHC as the primary testing methodology is supported by subset analysis of patients within the ToGA trial that showed trastuzumab plus chemotherapy improved overall survival in patients with high expression of HER2 protein (IHC 2+ and FISH positive or immunohistochemistry 3+) compared with patients with low expression of HER2 protein (IHC 0 or 1+ and FISH positive).

Interpretation

IHC results are reported as:

- Negative for HER2 Expression (0 or 1+)
- Equivocal for HER2 Expression (2+), and
- Positive for HER2 Expression (3+).

HER2 gene status is reported as either:

- *HER2* amplified or
- *HER2* non-amplified.

The *HER2* gene copy number is specified, and the *HER2*/CEP17 ratio is reported (in gastric and gastroesophageal carcinoma a normal non-amplified ratio is < 2.0).

Limitations

IHC is reliably performed in formalin-fixed paraffin-embedded tissues that have been processed in 10% neutral buffered formalin. Inconsistent results may be seen in specimens that were improperly fixed (incorrect fixative, delay in fixation, or abbreviated fixation duration) and in small biopsies that suffer from architectural distortion. Similarly, the FISH assay performs well using formalin-fixed paraffin-embedded tissue sections. Inconsistent FISH results are obtained in specimens processed in fixatives and preservatives that do not contain 10% neutral buffered formalin.

Methodology

IHC is performed utilizing the 4B5 rabbit monoclonal antibody (Ventana Medical Systems, Intl; Tucson, AZ). The interphase FISH probe assay (PathVysion, Abbott Molecular, Vysis, Des Plaines, IL) is a dual DNA probe assay containing a *HER2* probe that spans the entire *HER2* gene and a CEP 17 probe that hybridizes to the alpha satellite DNA located at the centromere of chromosome 17 (17p11.1-q11.1). With the inclusion of the CEP 17 probe, the relative copy number of the *HER2* gene can be determined and is expressed as the *HER2*/CEP17 ratio.

References

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Test Overview

Test Name	Erb-b2
Cross References and Aliases	Her 2 Neu Immunohistochemical Technique; CERB-B2
Methodology	Immunohistochemistry; HER2 FISH
Reference Range	Erb-b2: Refer to report.
Internal Specimen Requirements (Main Campus)	Tube/Container: See note. Note: Submit 10% buffered formalin-fixed tissue, formalin-fixed paraffin block, frozen tissue or unstained formalin-fixed paraffin slides. Indicate patient's name, CCF number and special request, (i.e., Erb-2). Label paraffin blocks with referring hospital name.
Internal Specimen Requirements (FHC/RMP)	Tube/Container: See note. Transport Temperature: Ambient. Note: Submit 10% buffered formalin-fixed tissue, formalin-fixed paraffin block, frozen tissue or unstained formalin-fixed paraffin slides. Indicate patient's name, CCF number and special request (i.e., Erb-2). Label paraffin blocks with referring hospital name.
External Specimen Requirements	Tube/Container: See note. Transport Temperature: Ambient. Note: Submit 10% buffered formalin-fixed tissue, formalin-fixed paraffin block, frozen tissue or unstained formalin-fixed paraffin slides. Indicate the patient's name, unique identifier and special request (i.e., Erb-2). Clearly label paraffin blocks with referring hospital name.
Alternate Specimen Requirements	Tube/Container: See note. Transport Temperature: Ambient. Note: Submit 10% buffered formalin-fixed tissue, formalin-fixed paraffin block, frozen tissue or unstained formalin-fixed paraffin slides. Indicate the patient's name, unique identifier and special request (i.e., Erb-2). Clearly label paraffin blocks with referring hospital name.
Special Information	Submit Surgical Pathology Requisition or accompany with an electronically generated test request. Interpretation by pathologist or request "Stain only – no interpretation." Paraffin blocks from referring hospitals must be labeled with the outside hospital name.
Billing Code	81012
CPT Code	88342

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