This requirement is applicable to both new and existing assays. If review of the initial validation/ verification does not meet the current standard, it must be supplemented and brought into compliance. If no records exist from the initial validation/verification, the assay must be fully revalidated/verified.

Evidence of Compliance:

✓   Records of validation/verification data including criteria for concordance

REFERENCES
1) Wolff AC, Somerfield MR, Dowsett M, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Guideline Update. *Arch Pathol Lab Med.* Published online June 7, 2023. doi: 10.5858/arpa.2023-0905-SA.
2) Fitzgibbons PL, Murphy DA, Hammond ME, Allred DC, Valenstein P. Recommendations for validating estrogen and progesterone receptor immunohistochemistry assays. *Arch Pathol Lab Med* 2010; 134:930-935.
3) Wolff AC, Hammond ME, Hicks DG, *et al.* Recommendations for human epidermal growth factor receptor 2 testing in breast cancer; American Society of Clinical Oncology/College of American Pathologists. *Arch Pathol Lab Med* 2014;138(2):241-256
4) Goldsmith JD, Troxell M, Roy-Chowdhuri S, et al. Principles of analytic validation of immunohistochemical assays: guideline update. *Arch Pathol Lab Med.* 2024. https://doi.org/10.5858/arpa.2023-0483-CP

CYP.04540   Estrogen Receptor and HER2 Testing in Breast Cancer Samples   Phase I

At least one tumor sample from all patients with invasive breast cancer (newly diagnosed, recurrent, or metastatic disease) is tested for estrogen receptors and HER2 (by IHC or ISH) if tissue is available.

REFERENCES
1) Wolff AC, Somerfield MR, Dowsett M, et al. Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Guideline Update. *Arch Pathol Lab Med.* Published online June 7, 2023. doi: 10.5858/arpa.2023-0905-SA.

CYP.04550   Fixation - HER2 and ER Breast Cancer Predictive Marker Testing   Phase I

If the laboratory assesses HER2 protein over-expression by immunocytochemistry, or estrogen receptor expression by immunocytochemistry for breast cancer predictive marker testing, the laboratory monitors cold ischemia time (one hour or less), if applicable, and appropriate specimen fixation time.

NOTE: The CAP strongly recommends that specimens subject to these tests be fixed in 10% neutral buffered formalin for at least six hours and up to 72 hours at room temperature. Specimens must be fully submerged in the optimal volume of formalin to achieve a formalin to specimen volume of 10:1 or higher, or if not feasible (eg, large specimens) at least 4:1. For cases with negative HER2 results by immunocytochemistry that were fixed outside these limits, confirmatory analysis by in situ hybridization is strongly recommended.

Both the time of removal of the tissue and the time of immersion of the tissue in fixative must be recorded and communicated from the submitting service to the processing laboratory.

Communication to clinical services of the need for appropriate information on cold ischemia time, fixative, and fixation time may be through memoranda, website, phone, face-to-face meetings, or other means. Information about fixative, fixation time, and cold ischemia time (if applicable) for each specimen must be recorded as part of the permanent specimen record in the pathology report. The laboratory must monitor for compliance and take corrective action as needed.

If specimens are fixed in a solution other than 10% neutral buffered formalin, the laboratory must perform a validation study showing that HER2 and ER results are concordant with results from formalin-fixed tissues.

Laboratories testing specimens obtained from another institution must have a policy that addresses cold ischemia time (if applicable) and time of fixation. Information on time of fixation may be obtained by appropriate questions on the laboratory's requisition form. If specimens have undergone any deviation from processing that may interfere with result interpretation, this must be annotated on the final report.