

SAMPLE IMMUNOHISTOCHEMISTRY REPORT

DIAGNOSIS:

ABC Hospital A08-03456

Breast, left, excision: Infiltrating ductal carcinoma, with the following immunophenotype:

1. **Positive** for estrogen and progesterone receptor expression
2. **Positive at 3+** for overexpression of HER2 oncogene protein (see comments).

COMMENTS:

Test Methodology: The method for HER2 immunohistochemistry (IHC) testing employed at PhenoPath Laboratories represents a validated modification of an FDA-approved method (HercepTest™). The IHC method incorporates the same A0485 rabbit polyclonal antibody employed in HercepTest™. **Scoring Methodology:** Results are scored as negative (either 0 or 1+ normalized immunostaining signal), positive (3+ normalized immunostaining signal), or equivocal (2+ normalized immunostaining signal). 3+ positivity is defined as uniform, strong signal on >30%. Scoring is performed as outlined in the ASCO-CAP Guidelines (Wolff AC et al., J Clin Oncol 25:118-45, 2007) with the addition of a "normalization" in which the level of signal on the non-neoplastic breast epithelium is subtracted from the score on the tumor. **Controls:** Separate 1+ and 3+ positive controls are employed for each run. **Quality Assurance:** PhenoPath Laboratories has in place a quality assurance program that includes daily case review conferences and ongoing concordance studies between IHC and FISH, to ensure high levels of interobserver and methodology concordance. **Validation of Assay:** Using this methodology, this laboratory has achieved an extremely high concordance rate between IHC and fluorescence in situ hybridization (FISH) assessment of HER2 status (Yaziji H et al., JAMA 291:1972-7, 2004; Gown AM et al., Breast Cancer Res Treatment 100:S218, 2006; Gown AM et al., Mod Pathol, 2008, in press). In the latter study of 6604 patients, a 99.2% concordance was demonstrated between negative (0 or 1+) HER2 IHC and non-amplified HER2 FISH, and a 94.7% concordance was demonstrated between positive (3+) HER2 IHC and amplified HER2 FISH.

MATERIAL RECEIVED

A1 = A08-03456, 2, 1 block

Fixative: Formalin

Duration of fixation >6 and <48 hours?: YES

CLIENT REQUEST / CLINICAL HISTORY

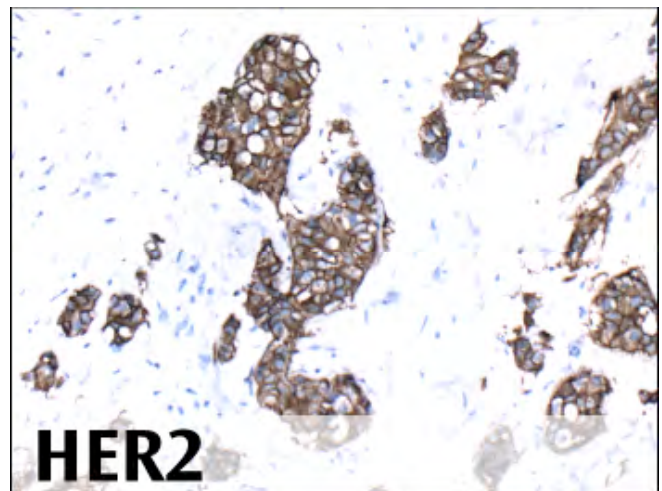
ER/PR/HER2 by IHC

IMMUNOHISTOCHEMICAL FINDINGS

Tissue sections (along with appropriate positive control) are incubated with the following antibodies. Localization is via a biotin-free, polymer-based immunoperoxidase technique according to an optimized protocol. The controls are reviewed for appropriate positive and negative reactivity and found to be satisfactory.

Block 2 (Surgery Date: 06/30/2008) - Breast, left (PP200800000 A1)

Target population: Tumor



Antibodies To	Clone	Result
Estrogen Receptor (§)	SP1	Positive [high level, 76-100%]
Progesterone receptor (§)	PgR 636	Positive [low level, 1-25%]
HER2 (§)	DAKO A0485 polyclonal	3+

• Electronically signed 07/02/2008: Allen M. Gown, M.D., Medical Director & Chief Pathologist

Per CMS regulations, the pathologist's signature above indicates that this case has been reviewed & the diagnosis made or confirmed by said pathologist.

NOTE: Some of the tests reported here may have been developed and performance characteristics determined by PhenoPath Laboratories. They have not been cleared or approved by the U.S. Food and Drug Administration (FDA). However, the FDA has determined that such clearance or approval is not necessary. Pursuant to the requirements of CLIA, this laboratory has established and verified the accuracy and precision of all tests, and additional information about these tests is available upon request. PhenoPath Laboratories is certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) as qualified to perform high complexity clinical laboratory testing.

Dr. Pathologist MD
ABC Hospital
Anywhere, WA 98102



PP2006-XXXXX-XX
PATIENT: Doe, Jane
DATE OF BIRTH: July 1, 1968
AGE: 40 GENDER: F
RECEIVED: July 1, 2008

EXPLANATORY NOTES ON ANTIBODIES AND PROBES

Estrogen receptor (ER) - Patients with ER-positive breast cancers have longer disease-free and overall survival, and a significantly higher likelihood of response to hormonal therapies such as tamoxifen, compared to those with ER-negative tumors (Bardou V-J, et al., J Clin Oncol 21:1973-9, 2003). This laboratory employs the anti-ER rabbit monoclonal antibody, SP1, which we have clinically validated in a long term retrospective study of over 4000 patients to better predict clinical outcome and tamoxifen response than the 1D5 mouse monoclonal antibody. The antibody and techniques employed in this laboratory identify an additional 5-10% of breast cancers as positive compared with the 1D5 antibody, and the long-term outcome of this patient cohort appears that of ER-positive breast cancer (Cheang M., et al., J Clin Oncol, 2006, in press). Results of hormone receptor studies are reported in a semiquantitative manner as "high level" positivity (corresponding to 76-100% of tumor cells positive), "intermediate level" positivity (corresponding to 26-75% of tumor cells positive), or "low level" positivity (corresponding to between 1 and 25% of the tumor cells positive). Published studies have suggested that even patients whose tumors show as few as 1-10% of tumor cells positive have significantly improved response to tamoxifen. Tumors with fewer than 1% of cells positive are reported as negative Harvey JM et al. J Clin Oncol 17:1474-81, 1999; Allred CA et al. Mod Pathol 11:155-68, 1998).