Biomarker Datasheet

Human HER2 OmniVUE™ Biomarker

HER2 (Human Epidermal Growth Factor Receptor 2) is a transmembrane tyrosine kinase receptor belonging to the EGFR family, involved in the regulation of cell growth and differentiation[1]. HER2 is overexpressed in certain cancers, most notably breast and gastric cancers, where it drives aggressive tumor cells proliferation, survival and tissue invasion [2]. HER2 protein overexpression or ERBB2 gene amplification are key predictive biomarker in oncology[3], guiding therapeutic decisions such as the use of targeted therapies like trastuzumab, pertuzumab, and Antibody-Drug Conjugates (ADCs) like trastuzumab deruxtecan. ADCs targeting HER2 have revolutionized the treatment of HER2-positive cancers and have led to the recent approval of the first tumor-agnostic HER2-directed therapy for previously treated patients with metastatic HER2-positive solid tumors[5]

Overview

Target	Other names	Isotype	Primary cell type	Subcellular location	Relevant Indications
HER2*	ERBB2, NEU, HER-2/neu	Rabbit IgG	Breast cancer cells, gastric cancer cells	Plasma Membrane	breast cancer, gastric cancer

*Clone available upon request

Quality Control

Each lot of antibody-barcode conjugate reagent is tested on the appropriate positive control tissue and reviewed by Ultivue's pathologists and image analysis experts to ensure expected staining pattern and positive signal intensity, through qualitative as well as quantitative analysis. Lot-to-lot consistency is evaluated and strictly maintained through quantitative comparison of a new lot of reagents with the predicate (previous lot) with an accepted variability of ±20% for positive signal intensity, which is at par with the current standards practiced in pathology.

Predicate Comparison

Serial sections of a graded breast cancer tumor TMA were stained with traditional chromogenic DAB using unconjugated antibodies and with the InSituPlex® (ISP) monoplex assay to demonstrate concordance between staining modalities (Figure 1).

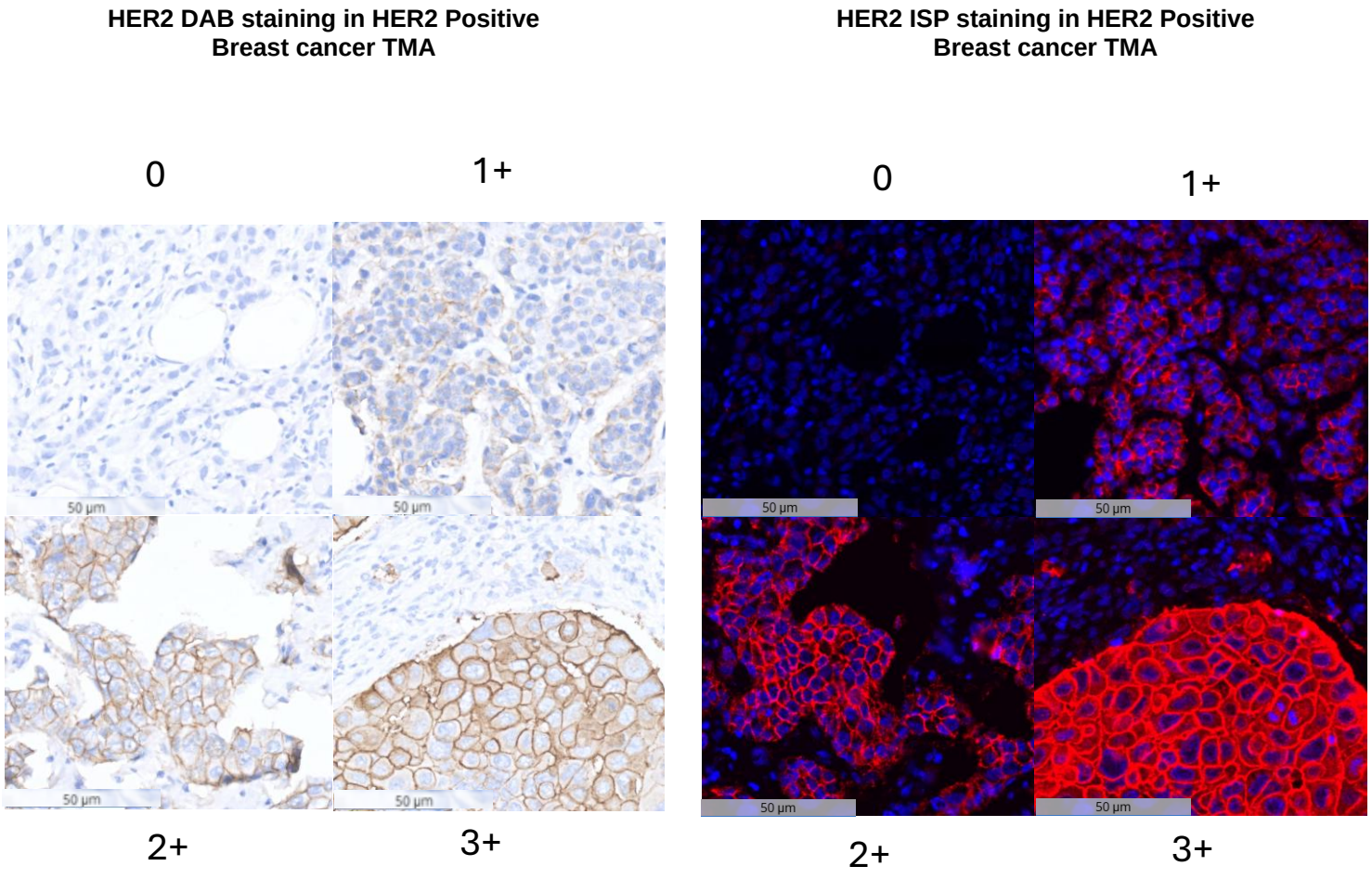


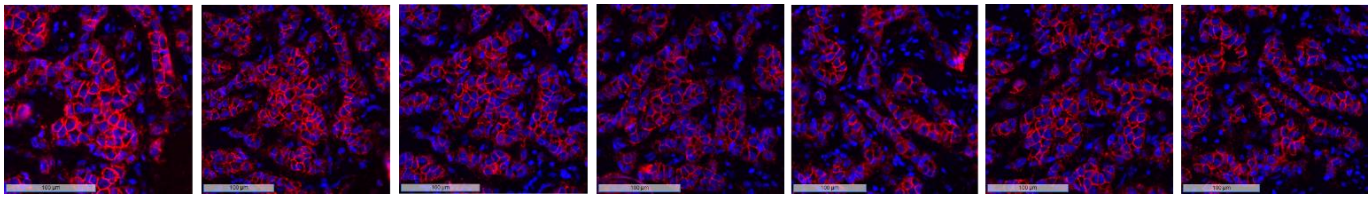
Figure 1: Comparison of unconjugated DAB and InSituPlex® monoplex assay in Breast CA tissue. Chromogenic DAB (left panel), fluorescent ISP staining (right panel).

Assay Precision Testing

Precision of the InSituPlex® monoplex assay was evaluated by assessing intra-run precision and repeatability across serial sections of the 2+ and 3+ cores of a graded breast cancer tumor TMA on the BOND RX autostainer by Leica Biosystems. For assessment of intra-run precision (Figure 2 and 3), 7 serial sections were stained within the same Bond RX run, imaged and analyzed in order to calculate the coefficient of variation (CV) per core for intra-run densities (number of positive cells/mm^2). Inter-day precision (Figure 4) was assessed by staining 3 serial sections across 3 different days on multiple Bond RX staining runs. Stained slides were imaged and analyzed to similarly calculate the CV per core for cell densities. Staining performance was found to be qualitatively and quantitatively concordant across all slides within a single run (intra-run precision testing) as demonstrated by CVs of positive cell density for each core. CVs for positive cell density were within the acceptance value of 30% for both the 2+ cores (P02: 9.9%, P05: 5.9%, P12: 5.1%) and the 3+ cores (P04: 6.0%, P07: 5.3%, P10: 4.7%). Similarly, staining performance demonstrated high concordance between slides stained across multiple runs. CVs for positive cell density remained within 30% for both the 2+ cores (P02: 9.2%, P05: 8.2%, P12: 10.0%) and the 3+ cores (P04: 8.3%, P07: 6.0%, P10: 2.2%) across the three independent runs.

We demonstrated excellent assay performance for both the 2+ and 3+ cores of the breast cancer TMA for which CVs were much lower than the highest accepted value of 20%[6]. To note, for tumor indications, the acceptance range is usually increased to 30% to accommodate possible tumor heterogeneity.

a



b

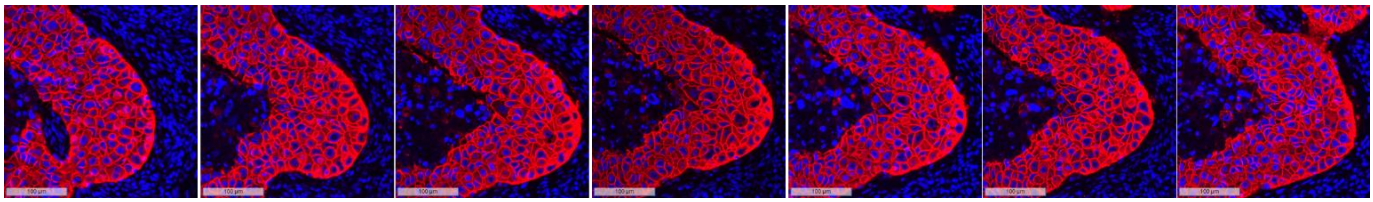


Figure 2: Intra-run precision of the InSituPlex® monoplex assay a. Representative images of serial sections of a 2+ TMA core. **b.** Representative images of serial sections of a 3+ TMA core.

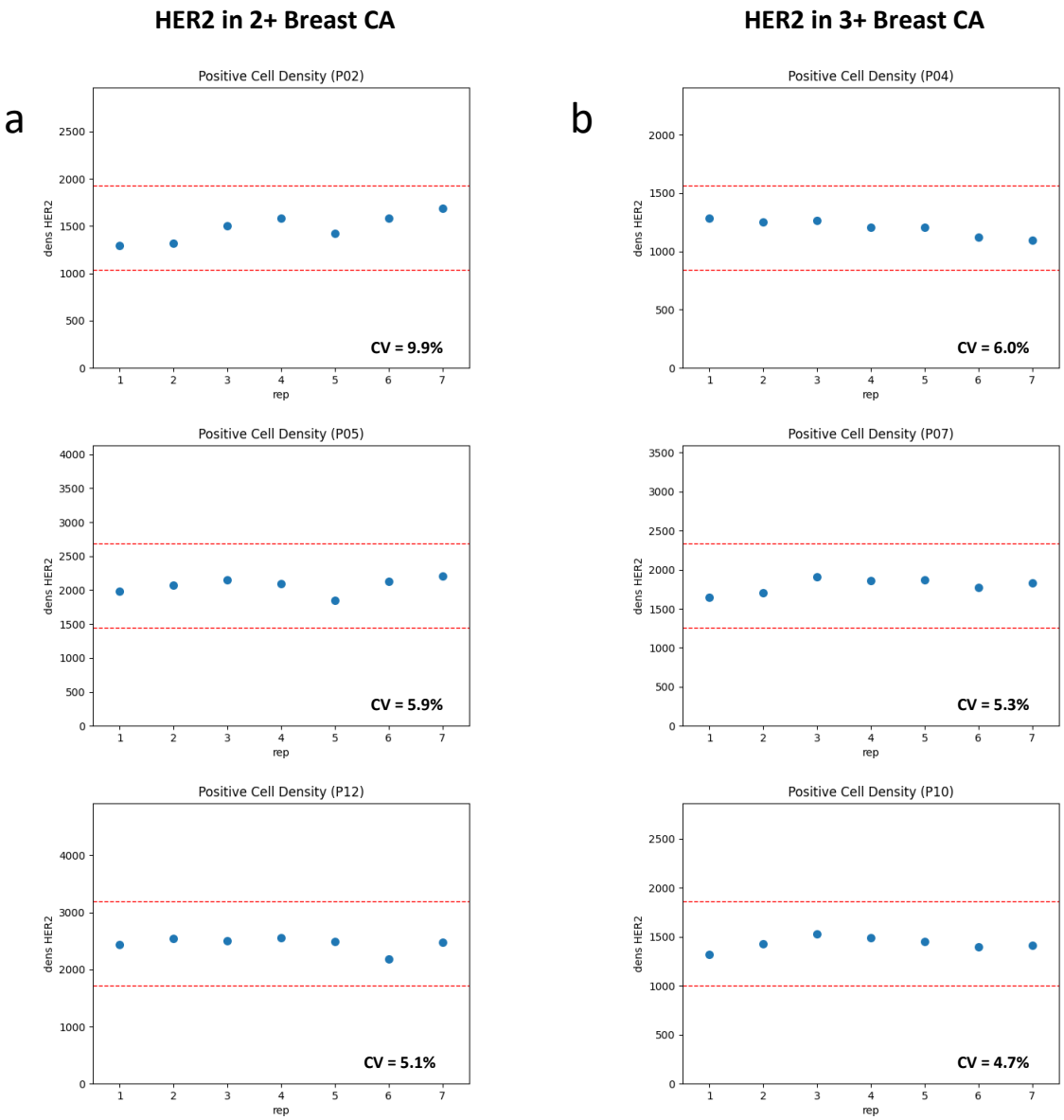


Figure 3: Intra-run precision of the InSituPlex® monoplex assay a. Number of positive cells/mm^2 per slide in each of three 2+ breast cancer core (P02, P05, P12). **b.** Number of positive cells/mm^2 per slide in each of the three 3+ breast cancer core (P04, P07, P10). For breast cancer tissue, thresholds (red dotted lines) mark +30% and -30% of the mean of all samples.

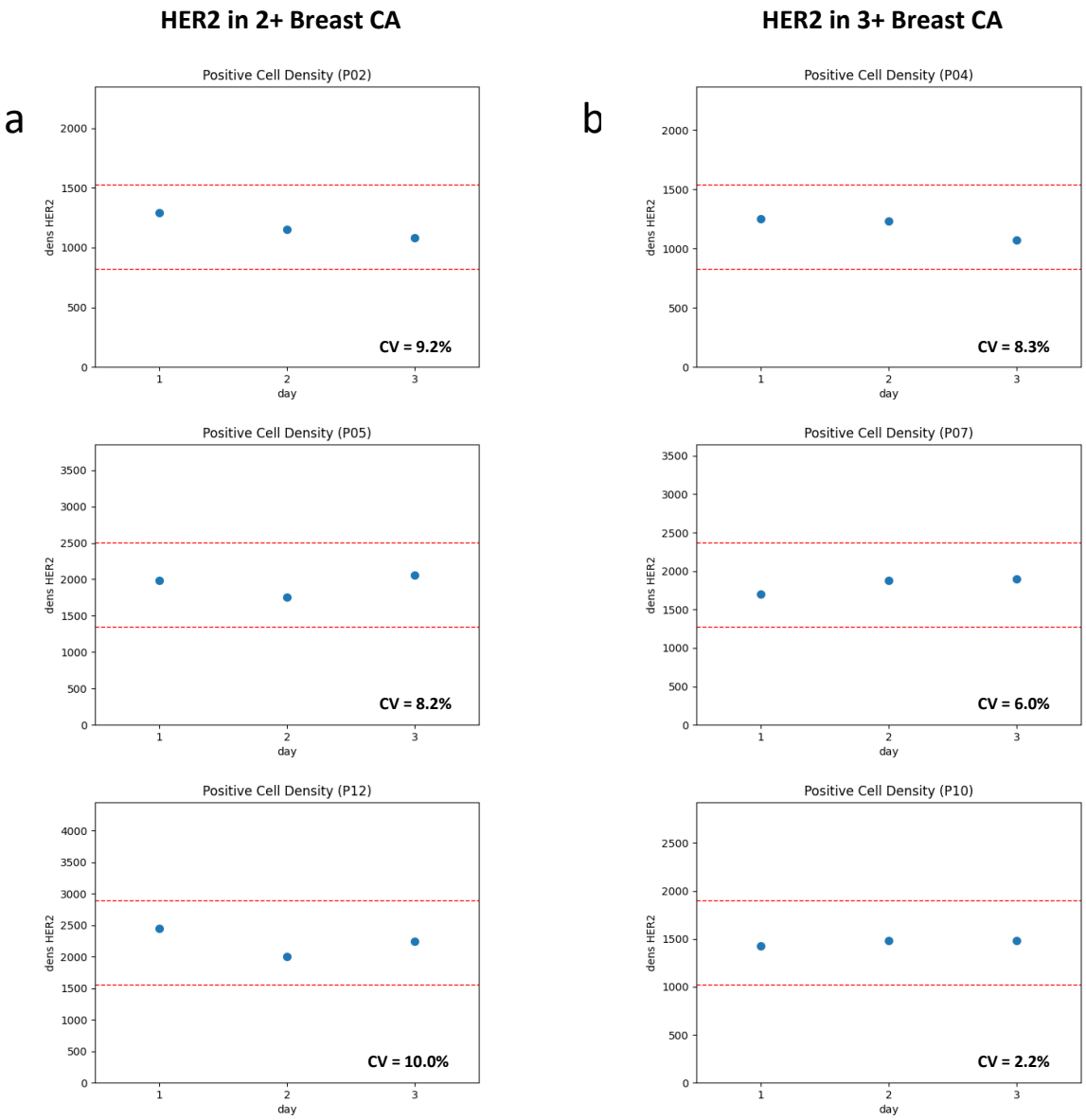


Figure 4: Inter-run precision of the InSituPlex® monoplex assay a. Number of positive cells/mm^2 per slide in each of the three 2+ breast cancer core (P02, P05, P12). **b.** Number of positive cells/mm^2 per slide in each of the three 3+ breast cancer core (P04, P07, P10). For breast cancer tissue, thresholds (red dotted lines) mark +30% and -30% of the mean of all samples.

References

1. Iqbal N, et al. Human Epidermal Growth Factor Receptor 2 (HER2) in Cancers: Overexpression and Therapeutic Implications*. Mol Biol Int*. 2014; 852748.
2. Baselga, J., & Swain, S. M. (2009). "Novel anticancer agents: the expanding role of HER2 inhibitors." *Nature Reviews Cancer, 9(7)*, 463-475.
3. Pillai R, et al. HER2 mutations in lung adenocarcinomas: A report from the Lung Cancer Mutation Consortium. *Cancer*. 2017;1;123(21): 4099-4105.
4. Verma, S., Miles, D., Gianni, L., et al. (2012). "Trastuzumab emtansine for HER2-positive advanced breast cancer." *New England Journal of Medicine, 367(19)*, 1783-1791.
5. FDA grants accelerated approval to fam-trastuzumab deruxtecan-nxki for unresectable or metastatic HER2-positive solid tumors 2024 https://www.fda.gov/drugs/resources-information-approved-drugs/fda-grants-accelerated-approval-fam-trastuzumab-deruxtecan-nxki-unresectable-or-metastatic-her2
6. Characterizing Intra-Tumor and Inter-Tumor Variability of Immune Cell Infiltrates in Murine Syngeneic Tumors Mojtahedzadeh, Sepideh et al. The American Journal of Pathology, Volume 191, Issue 12, 2133 - 2146