

## HER2 Expression is Associated with Altered Membrane Morphology in Prostate Cancer

Christina Matalaka<sup>1,2</sup>, Marcelina Goldstein<sup>1,2</sup>, Lauren Smith<sup>1,2</sup>, Christopher Chan<sup>1,2</sup>, Emily C. Williams<sup>1,2</sup>, Maneesh Jain<sup>2,3,4</sup>, Inhee Chung<sup>1,2</sup>, and Maho Shibata<sup>1,2,4</sup>

<sup>1</sup>Department of Anatomy and Cell Biology, The George Washington University School of Medicine and Health Sciences, Washington, DC, USA

<sup>2</sup>The George Washington University Cancer Center, The George Washington University School of Medicine and Health Sciences, Washington, DC, USA

<sup>3</sup>Section of Hematology and Oncology, Washington DC VA Medical Center, Washington, DC, USA

<sup>4</sup>The Edward P. Evans Precision Oncology Center of Excellence at the DC VA Medical Center and GW Cancer Center, Washington, DC, USA

There remains a need to understand how human epidermal growth factor receptor 2 (HER2) expression promotes prostate cancer progression. Unlike in breast cancers, HER2 gene amplification is rare in prostate cancers, and HER2 expression is typically low and heterogeneous. However, when scored using a modified prostate-specific HER2 immunohistochemistry scoring system adapted from gastrointestinal cancers, tumors from approximately 60% of patients with prostate cancer scored positive for membrane HER2 expression (Estephan et al. 2023). Trastuzumab deruxtecan (T-DXd), an antibody-drug conjugate targeting HER2, improves progression-free and overall survival for patients with HER2-low breast cancers. Based on the rationale that HER2-low prostate cancer patients may benefit similarly, a phase II clinical trial evaluating the efficacy and safety of T-DXd for HER2-positive metastatic castration-resistant prostate cancer is ongoing (Castrate Resistant Prostate Cancer Enhertu Therapy, CaRPET Clinical trial, NCT06610825).

HER2 overexpression in breast cancer cells alters membrane morphology, promoting the formation of finger-like membrane structures that can be identified by electron microscopy (Chung et al. 2016). These membrane deformations reduce cell contacts with neighboring cells and promote cell motility. We therefore hypothesized that similar cell membrane deformations occur in HER2-expressing prostate cancers. In this study, we evaluated membrane HER2 expression in wild-type and prostate cancer cells using two genetically-engineered mouse models of prostate cancer and prostate tumor organoids generated from these mice. We then performed scanning electron microscopy on prostate organoids to assess membrane morphology. We detected membrane HER2 expression in prostate tumor cells from genetically engineered mouse models of prostate cancer and found that HER2 membrane expression was maintained in tumor organoids. While HER2 expression was limited in wild-type prostates, we observed membrane HER2 expression in luminal cells in the proximal prostate and in wild-type organoids. Analysis of scanning electron microscopy images revealed that tumor organoids with higher membrane HER2 expression had increased membrane deformations and protrusions, as well as increased intercellular distances between cells. Overall, our findings suggest a conserved role for membrane HER2 expression in promoting changes in membrane morphology in breast and prostate cancers.

**Funding Acknowledgments:** This work was supported by National Institutes of Health (NIH) grant R00CA194287, the Prostate Cancer Foundation and Edward P. Evans Precision Oncology Center of Excellence at the DC VA Medical Center and GW Cancer Center, and the GW Cancer Center Katzen Research Program.

**Conflicts of Interest:** The authors declare no conflicts of interest.

### References:

Chung I, Reichelt M, Shao L, Akita RW, Koeppen H, Rangell L, Schaefer G, Mellman I, Sliwkowski MX. 2016. High cell-surface density of HER2 deforms cell membranes. *Nature communications* 7: 12742.

Estephan F, Lap CJ, Banagan J, Antonio M, Liu S, Diao G, Rozalen AZ, Rajendran R, Krasnow S, Subrahmanyam R, Nava VE, Jain M. 2023. The prevalence and clinical significance of HER2 expression in prostate adenocarcinoma. *Ann Diagn Pathol* 67: 152219.