

# Validating and Evaluating Expression of New Tumor Targets Across Cancers: Protein X as a Potential Target for Cancer Drug Development

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Lung and ovarian cancers continue to be leading causes of cancer deaths in the United States, and refining the treatment of these cancers is needed to improve patient survival. The goal of this research project was to develop a methodology to identify protein biomarkers as potential targets for refined cancer treatment. This novel methodology combined an immunohistochemistry assay with a prevalence study. The immunohistochemistry assay was developed with an antibody selection test to confirm antibody specificity, a titration test to optimize antibody concentration, and an antigen retrieval test to optimize epitope-antibody binding. The prevalence study involved a robust assay, digital quantitative image analysis, and data analysis. With this methodology, the important immune system protein, Protein X, was quantified and evaluated across two indications of cancer: non-small cell lung cancer (NSCLC) and ovarian cancer. Protein X expression was quantified and evaluated across 594 samples from NSCLC and ovarian cancer patients. Data analysis demonstrated that Protein X was significantly expressed on NSCLC ( $p < 0.0001$ ) compared to its expression in normal tissue, and significantly expressed on one major NSCLC subtype, squamous cell lung cancer ( $p < 0.003$ ). Protein X was not significantly expressed in ovarian cancer. The findings from this project indicate that Protein X is a potential biomarker and refined treatment target for NSCLC. Furthermore, the optimized immunohistochemistry assay developed in this study is applicable to identify other novel cancer biomarkers for precision drug development in a standardized manner.