Artificial Intelligence (AI)-Driven Quantification of p53 Expression Improves Prognostic Stratification in Prostate Cancer

Keith Kyewalabye[1], Palak Patel[1], Mackenzie Bennett[1], Amber Bixby[1], James Corines[1], Neeru Gandotra[1], Michel R. Nasr[1], Ola El-Zammar[1], Alina Basnet[2,3] and Tamara Jamaspishvili[1]

[1]Department of Pathology and Laboratory Medicine, SUNY Upstate Medical University, NY, USA
[2]Department of Medicine, SUNY Upstate Medical University, NY, USA
[3]Renzi Cancer Center, Guthrie Cortland Medical Center, NY, USA

Background: The tumor suppressor gene TP53 is one of the most frequently mutated genes in advanced prostate cancer, typically assessed by next-generation sequencing (NGS). Immunohistochemistry (IHC) serves as a potential surrogate for missense mutations that cause p53 nuclear accumulation. However, the accuracy and reproducibility of IHC-based p53 assessment are limited by subjective manual scoring, lack of standardized thresholds, and marked intratumoral heterogeneity.

Methods: To address these challenges, we developed an artificial intelligence (AI)-based quantitative algorithm using the Visiopharm image analysis platform to measure p53 expression on IHC whole-slide images and assess associations with survival outcomes. An institutional cohort of 383 patients with intermediate- and high-risk prostate cancer (2011-2021) was analyzed. Representative FFPE blocks were selected per patient, including 253 biopsies, 129 prostatectomies, and 1 TURP specimen. p53 IHC was performed in a CLIA-certified laboratory using a mouse monoclonal antibody (D0-7, Cell Marque) on the Ventana Benchmark ULTRA system with OptiView DAB detection (Roche/Ventana). Slides were scanned at 20× magnification (Aperio Leica AT2 Dx). Convolutional neural network (CNN)-based algorithms were trained to automatically detect cancer regions and quantify p53 staining intensity (0, 1+, 2+ 3+) and the proportion (%) of positive tumor nuclei. AI-derived metrics were correlated with progression-free (PFS), biochemical recurrence-free (BCRFS), and metastasis-free survival (MFS) using Kaplan-Meier analysis.

Results: Tumors with high (3+) nuclear intensity (261/383; 68.1%) exhibited significantly shorter PFS (median 40 vs. 105.2 months), BCRFS (53.9 vs. 122.4 months), and MFS (185.6 vs. 234.3 months; all $p < 0.05$) compared with those without 3+ intensity staining. Among 3+ cases, lower fractions (<1%) of p53-positive nuclei were associated with progressively shorter PFS and BCRFS (12.3, 26.5, and 54.8 months for <1%, 1-5%, and ≥5%, respectively). When tumors were stratified only by the percentage of p53-positive nuclei (<1%, 1-5%, ≥5%), ignoring intensity, reduced PFS was again observed for lower-level or focal expression (34.0, 36.8, and 62.5 months; $p < 0.05$), suggesting that subclonal TP53 alterations may be detectable by AI-based IHC quantification. In 13 sequenced cases, low (<1%) p53-positive fractions corresponded to TP53 copy number loss or frameshift mutations, supporting that focal p53 expression may capture loss-of-function events.

Conclusions: Strong (3+) p53 nuclear staining, even when limited to a small fraction of tumor cells, correlates with poor outcomes in prostate cancer, likely reflecting subclonal TP53 mutations. Very low (<1%) p53 positivity likely indicates frameshift or loss-of-function variants, also linked to poor prognosis. Although validation with sequencing is warranted, these findings underscore the utility of AI-assisted IHC quantification in refining biomarker assessment and capturing clinically relevant clonal and spatial heterogeneity.

Funding Acknowledgements: This study is supported by Prostate Cancer Foundation (Young Investigator Award)

Conflict of Interest Statement: The authors declare no conflicts of interest.