Club-like cells and PIGR expression are associated with inflammatory high-risk, localized prostate cancer

Anson Ku[1*], Rong Rong Huang[2*], Sumeyra Kartel[1], Kayla Heyward[1], Ross Lake[3], Rosina Lis[1], Nikil Pramod[1], Sara Dawood[1], Shana Trostel[1], Anna Baj[1], Etyan Ruppin[4], Scott Wilkinson[1], John Fenimore[1], Huihui Ye[5+,], Adam Sowalsky[1+]

Affiliations:

1. Genitourinary Malignancy Branch (GMB) at the National Cancer Institute (NCI), NIH, Bethesda, MD 20892, USA
2. Department of pathology and Laboratory Medicine, University of California, Los Angeles (UCLA), Los Angeles, CA 90095, USA
3. Laboratory of Cancer Biology and Genetics at the National Cancer Institute (NCI), NIH, Bethesda, MD 20892, USA
4. Cancer Data Science Laboratory at the National Cancer Institute (NCI), NIH, Bethesda, MD 20892, USA
5. Pathology and Laboratory Medicine at Cedars-Sinai, Los Angeles, CA 90048, USA

Background

Immunotherapy such has checkpoint inhibitors has transformed the treatment of many solid malignancies. Despite advances in the mechanistic understanding of treatment resistance, high-risk, localized prostate cancer are largely unresponsive towards immunotherapy. Interestingly, various publications have observed proportion of localized prostate cancer to be immunogenic that are unlikely to harbor microsatellite instability high or mismatch repair deficiency. To better understand the mechanism of immunogenicity, we compared prostate specimens with high or low tumor infiltrating lymphocytes (TILs) using spatial transcriptomics in order identify the mechanism immune recruitment.

Methods

Patient cohort

Radical prostatectomy specimens from untreated, high-risk, localized prostate cancer patients obtained from the UCLA biobank were screened for intra-tumoral T-cell infiltration (TILs) by multiplex immunohistochemistry for immune and tumor cells. Four samples were selected and profiled using the 10X Visium platform for FFPE specimens. Validation was performed using OPAL multiplex immune-fluorescence (mxIF) staining for CD4, CD8A, CD68, PIGR (marker of club cells), and PSA on 18 specimens from a separate cohort of untreated, intermediate to high-risk localized prostate cancer.

Statistics

Spatial analyses were performed on Visium data after clustering and spatial deconvolution to identify cell types. Co-occupancy analysis was used to identify T-cell colocalization partners. Cell segmentation was performed on mxIF images using HALO system. Spatial regression was performed on all stained images. All analysis were performed in R version 4.2.0.

Results

Spatial transcriptomic analysis from four patients identified myeloid cells and club cells to preferentially colocalize with T-cells. In areas of co-enrichment, interferon-gamma pathway and MHC-I were upregulated. MxIF confirmed the co-enrichment of T-cells with PIGR+ cells with CD4+ as the dominant population of T-cells. We observed PIGR to be expressed predominantly by benign or atrophic epithelium. Cancer cells can also express PIGR. Tumor PIGR are spatially associated with TILs. In-addition, cribriform morphology and PTEN loss were associated with increased TILs. Single-nuclei ATAC seq analysis revealed AP-1 transcription factor associated with club-cells and PIGR expression.

Conclusions
We defined a subset of immunogenic localized prostate cancer that expresses PIGR either in benign or tumor epithelium. Epithelial expression of PIGR is associated with TILs, suggesting a non-canonical route for immune recruitment and activation.

Funding Acknowledgements
Portions of this work utilized the computational resources of the NIH HPC Biowulf cluster; Prostate Cancer Foundation (Young Investigator Awards to A.K. and S.W.); Department of Defense Prostate Cancer Research Program (W81XWH-22-1-0067 to A.T.K. and W81XWH-19-1-0712 to S.W.); Intramural Research Program of the NIH, National Cancer Institute.

Conflict of Interest
None