

Decoding Stromal Heterogeneity across BPH Phenotypes

Diya Binoy Joseph¹, Gervaise H Henry¹, Alicia Malewska¹, Jeffrey C Reese², Ryan C Hutchinson¹, Claus G Roehrborn¹, Douglas W Strand¹



¹Department of Urology, UT Southwestern Medical Center, Dallas, TX 75390, USA

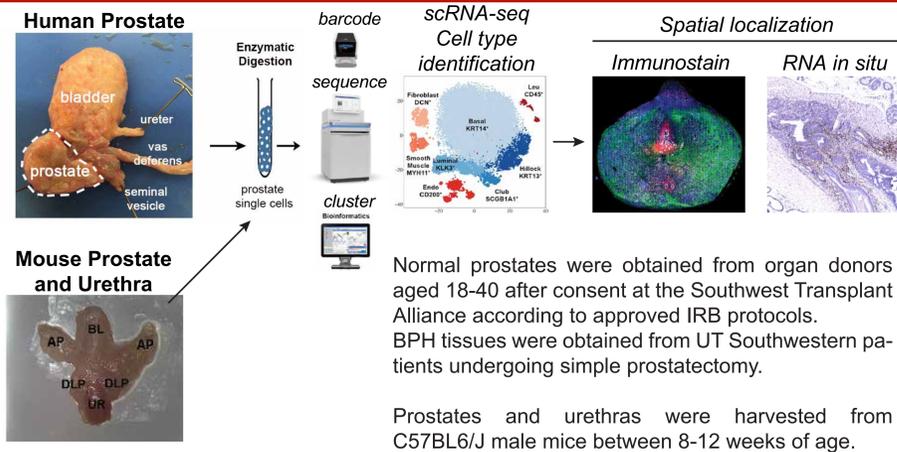
²Southwest Transplant Alliance, Dallas, TX 75231, USA



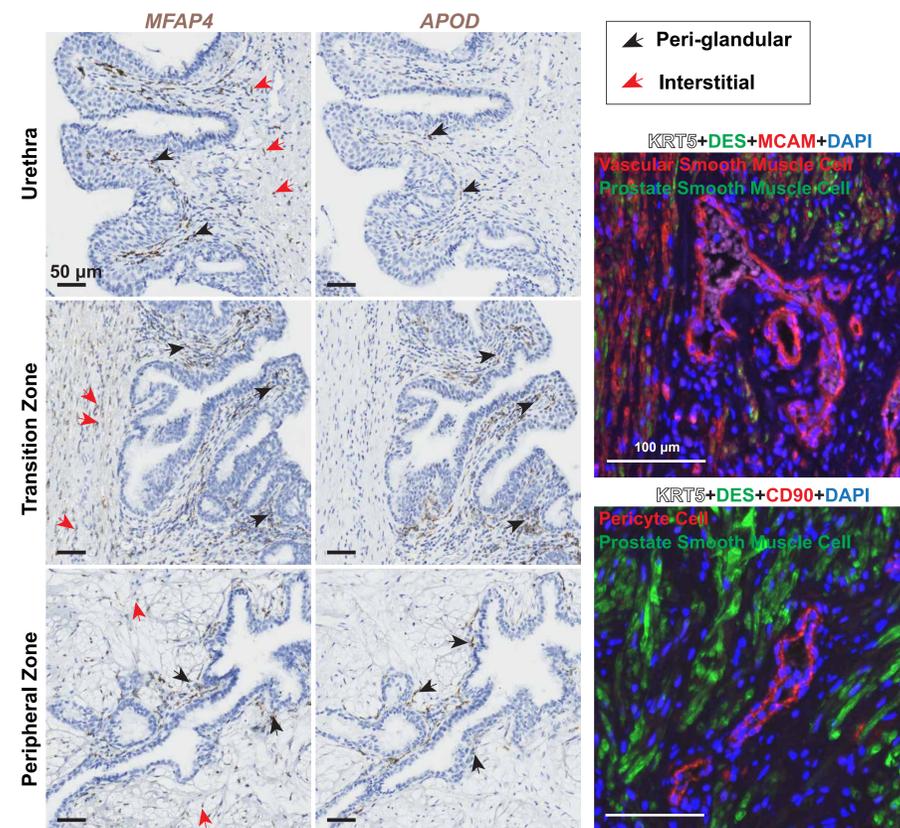
Introduction

Benign Prostatic Hyperplasia (BPH) is a non-malignant enlargement of the prostate that occurs with aging and is associated with Lower Urinary Tract Symptoms (LUTS). Therapeutic options often fail, necessitating surgical resection of the prostate. The phenotypic and cellular heterogeneity of BPH is thought to contribute to treatment resistance. BPH patients present with multiple nodules grouped around the prostatic urethra in the transition zone. The composition of these nodules vary with some being solely comprised of stromal cells and others containing a mixture of stromal and epithelial cells. In addition, some patients present with a band of fibrotic tissue around the prostatic urethra that we term as peri-urethral fibrosis. Here, we describe stromal cell heterogeneity in the normal human prostate and across the different BPH phenotypes. We carried out single cell RNA-sequencing on normal and BPH prostates and analysed fibromuscular stroma cells. We identified two fibroblast clusters in the human which localized to different spatial regions in the prostate. In addition, BPH phenotypes showed different compositions of these fibroblasts. We also carried out single cell RNA sequencing of the mouse prostate and urethra and identified the spatial localization of three different fibroblast sub-types.

Approach

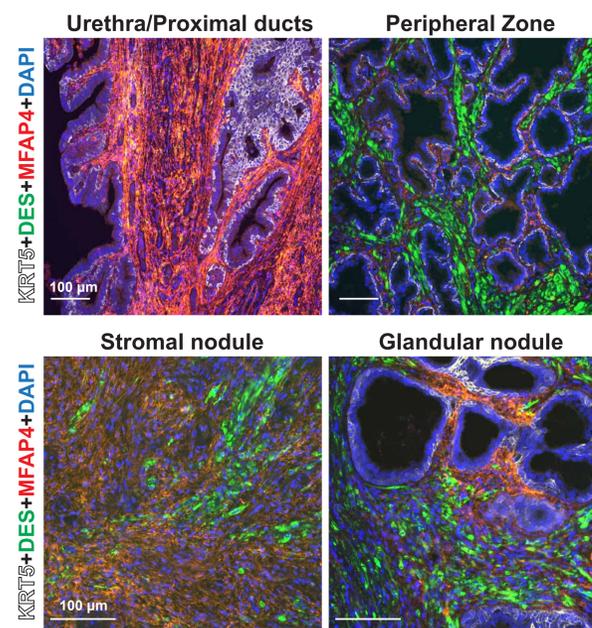


APOD+ fibroblasts localize to the peri-glandular region



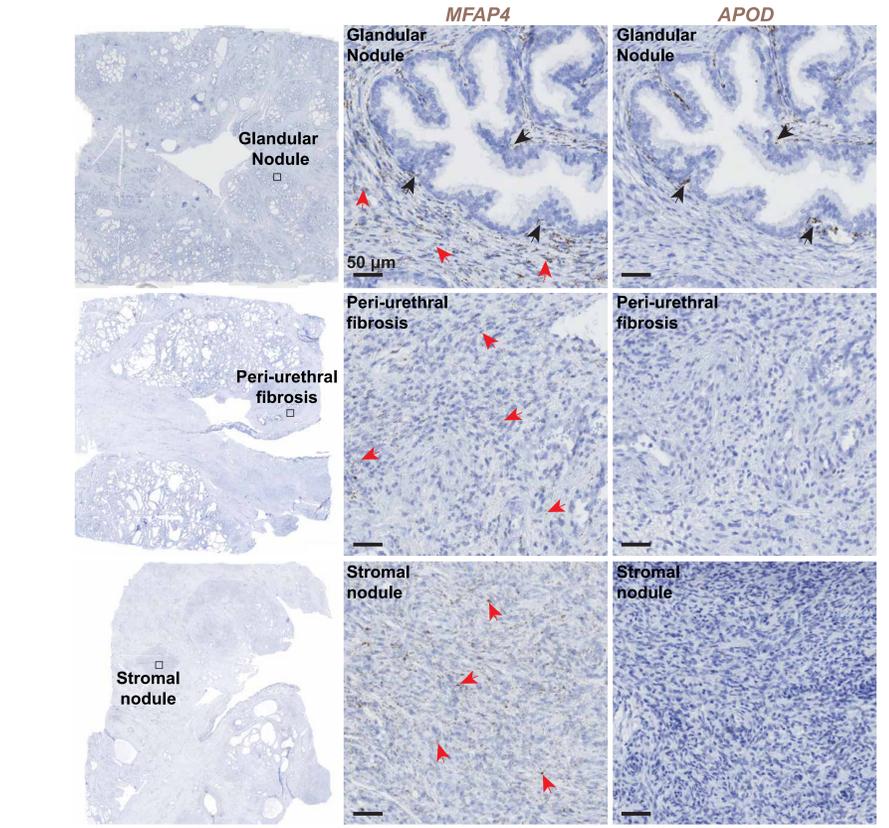
RNA in situ hybridization was used to localize mRNA expression of MFAP4 and APOD in different anatomical regions of the normal human prostate. Staining on adjacent serial sections indicate that APOD+ fibroblasts also express MFAP4, corroborating the single cell RNA-seq analysis. APOD+ fibroblasts are restricted to the peri-glandular regions while MFAP4+ fibroblasts can also be observed in the interstitial spaces between epithelial structures. Both fibroblast types are found in all anatomical regions of the prostate including the prostatic urethra. Various smooth muscle cell types can be observed in the prostate including vascular smooth muscle/pericytes (labeled by MCAM, CD90) and prostatic smooth muscle cells (labeled by Desmin).

Regional and phenotypic differences in fibroblast density



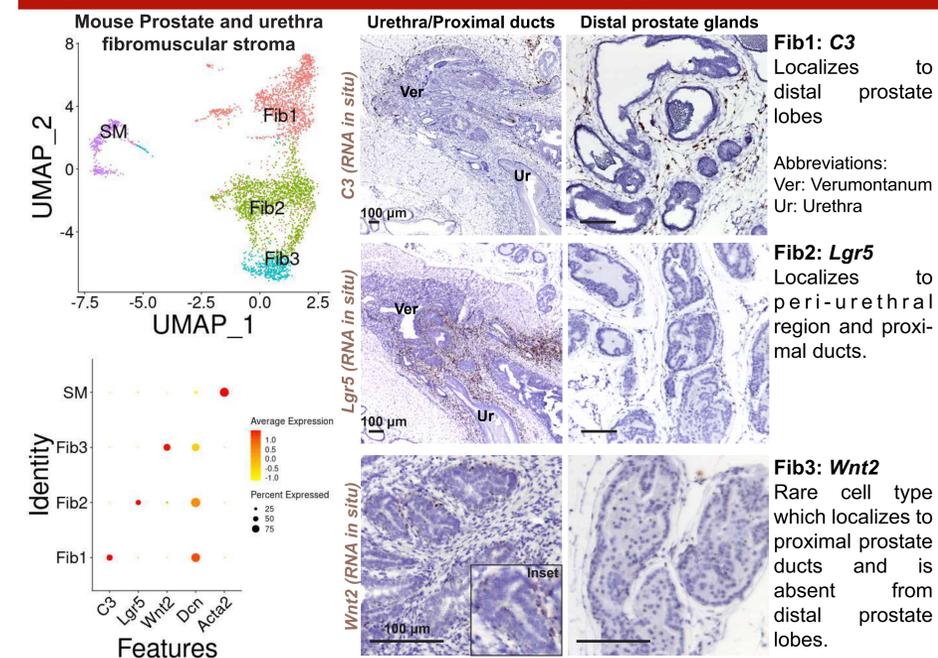
The stromal regions surrounding the prostatic urethra and peri-urethral ducts in the transition zone are rich in fibroblasts. The stromal composition of the peripheral zone is high in prostatic smooth muscle. Fibroblasts are present in thin bands around individual ducts and scattered in the interstitium. Stromal nodules are often found in the transition zone of BPH patients. These nodules are mostly comprised of fibroblasts with a few wisps of smooth muscle. Stromal nodules resemble the peri-urethral stroma in the normal prostate and could represent a localized expansion of fibroblasts due to inflammatory or other stimuli. In contrast, epithelia in glandular nodules have fibroblast and smooth muscle distribution resembling the peripheral zone of the normal prostate.

Fibroblast heterogeneity across BPH phenotypes

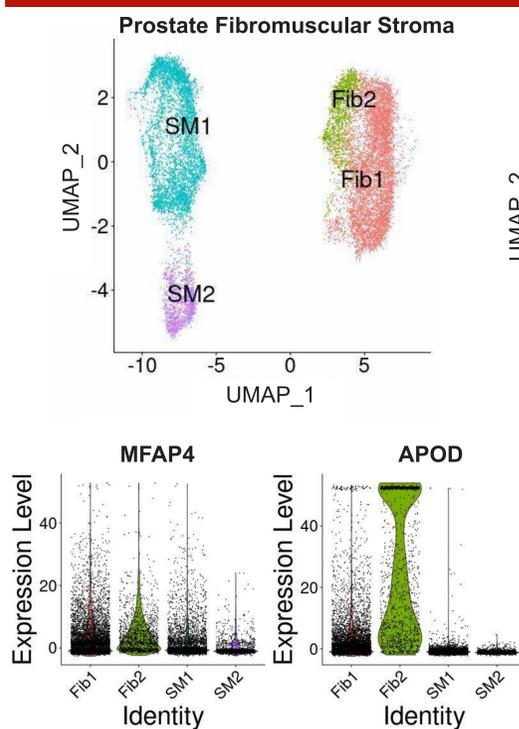


RNA in situ hybridization was used to localize mRNA expression of MFAP4 and APOD in different BPH histologic phenotypes. APOD+ fibroblasts are absent from stromal nodules and regions of peri-urethral fibrosis.

Spatial distribution of fibroblast sub-types in the mouse



Stromal heterogeneity in the human prostate



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