Susceptibility-associated genetic variation in NEDD9 contributes to prostate cancer progression

Background: In the US, African American (AA) patients show higher prostate cancer (PCa) incidents and worse treatment outcomes in comparison to European Americans (EA). This PCa disparity is largely contributed by socioeconomic factors, but race-associated genetic variations may also play a role. From GWAS analyses, we identified a panel of PCa risk-associated single nucleotide polymorphisms (SNPs) with significantly different allele frequencies in AA versus EA. The top-ranked SNP, rs4713266, is located at an intronic region of the NEDD9 gene and the risk allele frequency is significantly higher in AA. NEDD9 encodes for a focal adhesion protein that is phosphorylated by FAK and Src and acts as a signaling hub to regulate multiple downstream pathways. Clinically, NEDD9 amplification was found in castration-resistant PCa (~3%) and neuroendocrine PCa (~15%). The chromatin region containing this SNP is highly enriched for enhancer marks, indicating it may contain a putative enhancer (we named it NEDD9-Int1Enh) to drive NEDD9 transcription.

Methods: To determine the role of rs4713266 in regulating NEDD9 expression, we performed: (1) CRISPRa to determine if NEDD9-Int1Enh mediates NEDD9 transcription; (2) reporter assays to determine if the nucleotide variation of this SNP can alter binding of specific transcription factors; (3) CRISPR editing to modify the nucleotide at the SNP to determine the effects on NEDD9 expression. To determine NEDD9 function in PCa, we silenced NEDD9 in VCaP cells and then assessed its effects on the global gene profile using RNA-seq and on PCa tumor growth and metastasis using in vivo and in vitro models, such as zebrafish embryo injection.

Results: We found that forced activation of NEDD9-Int1Enh can increase NEDD9 expression and modifying the nucleotide of the SNP altered NEDD9 expression by interacting with distinct transcription factors such as ERG (non-risk) and NANOG (risk). Moreover, RNA-seq results revealed that NEDD9 may promote several oncogenic pathways, including epithelial-mesenchymal transition (EMT), JAK/STAT3, and KRAS signaling pathways. Indeed, we show that NEDD9 silencing decreased PCa tumor growth and metastasis in vitro and in vivo.

Conclusion: Our study indicates a critical role of rs4713266 on modulating NEDD9 expression in PCa of different racial subgroups. We also demonstrate strong oncogenic activities of NEDD9 in promoting PCa progression and metastasis. Together, our study provides novel insights into genetic mechanisms driving PCa racial disparities.