

Targeting the PTGES3-AR axis in Advanced Prostate Cancer

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Background:

The androgen receptor (AR) is a central driver of prostate cancer (PCa). While AR-targeted therapies provide survival benefit, most metastatic castration-resistant PCa (mCRPC) ultimately relapses through reactivation of AR signaling. To discover new therapeutic strategies, we developed live-cell endogenous AR fluorescent reporters and conducted genome-wide CRISPRi screens. These revealed prostaglandin E synthase 3 (PTGES3) as a top hit. PTGES3 loss markedly reduced AR protein, suppressed AR transcriptional programs, and impaired proliferation of AR-driven PCa models. Clinically, PTGES3 expression is elevated in advanced disease and associated with resistance to AR-directed therapy.

Methods and Results:

Mechanistically, we found PTGES3 localizes to both the cytoplasm and nucleus, with nuclear PTGES3 enriched in aggressive tumors. PTGES3 physically interacts with AR in the nucleus, binding to AR's DNA- and ligand-binding domains. Recombinant protein studies confirmed direct PTGES3-AR interaction, which enhanced AR's ability to bind canonical androgen response elements (AREs) and stabilized AR splice variant 7. Overexpression of PTGES3 augmented AR transcriptional activity, while knockdown of PTGES3 phenocopied AR loss: genome-wide ChIP-seq revealed >85% reduction of AR occupancy, and ATAC-seq showed broad loss of chromatin accessibility at hallmark AREs, including KLK3. These findings define PTGES3 as a nuclear co-factor that reinforces AR DNA binding and transcriptional output, independent of HSP90, and essential for sustaining AR signaling in mCRPC.

To translate these findings, we performed a disulfide tethering fragment screen targeting PTGES3, identifying covalent binders that disrupted PTGES3-AR interaction. Medicinal optimization yielded rac-18, a first-in-class covalent PTGES3 inhibitor. Rac-18 covalently modifies PTGES3, stabilizes its fold, and blocks AR binding. In AR-dependent cell lines, rac-18 reduced AR protein levels, suppressed AR transcriptional programs (RNA-seq), and decreased chromatin accessibility at AR-regulated loci (ATAC-seq). Rac-18 selectively impaired proliferation of AR-driven but not AR-independent PCa models. In vivo, rac-18 treatment significantly reduced growth of 22Rv1 xenografts, decreased tumor AR levels, and repressed AR target gene expression.

Conclusions:

We identify PTGES3 as a nuclear AR co-factor that stabilizes AR, enhances DNA binding, and promotes chromatin accessibility at androgen-responsive loci. Targeting this interaction with rac-18 disrupts AR signaling and selectively suppresses tumor growth in AR-driven models. These findings establish PTGES3 as a previously unrecognized vulnerability in prostate cancer and rac-18 as the first covalent inhibitor of this pathway, offering a promising therapeutic strategy to overcome resistance to current AR-directed therapies in mCRPC.

Acknowledgements:

This study is supported by Prostate Cancer Foundation Young Investigator Award and the UCSF Prostate Cancer Program Pilot Research Awards to HL. This study is funded by Prostate Cancer Foundation Challenge Awards and UCSF Benioff Initiative for Prostate Cancer Research award.