

Activation of P-TEFb by Androgen Receptor-Regulated Enhancer RNAs in Castration-Resistant Prostate Cancer

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Background: Androgen deprivation therapy has long been the mainstay of treatment for advanced prostate cancer, but tumors inevitably become castration-resistant prostate cancer (CRPC). Increasing evidence suggests that persistent androgen receptor (AR) signaling plays an essential role in development of hormone therapy resistance. It has been shown recently that enhancer RNAs (eRNAs) is widely transcribed from cell lineage-specific enhancers. Mechanistic studies show that in a locus-specific manner, eRNAs act *in cis* to stabilize enhancer-promoter looping, establish chromatin accessibility or facilitate release of the negative elongation factor (NELF) complex. Estrogen receptor (ER)- or AR-regulated eRNAs also act *in trans* to regulate gene expression in hormone-responsive cells, but the effects appear to be relatively infrequent in the cell types examined. To date, however, the function and disease relevance of AR-bound enhancer RNAs (AR-eRNAs) remain poorly understood.

Methods: High throughput AR ChIP-seq and strand-specific RNA-seq approaches were employed to identify AR-regulated enhancer RNAs (AR-eRNAs). 3C assays were used to verify the enhancers that express AR-eRNAs. Small interference RNAs and TALEN-mediated gene deletion were utilized to assess the functions of AR-eRNAs in gene transcription regulation and prostate cancer cell growth.

Results: We identified a group of AR-regulated enhancer RNAs (e.g. PSA eRNA) that were upregulated in CRPC cells, patient-derived xenografts (PDX) and patient tissues. PSA eRNA bound to CYCLIN T1, activated P-TEFb and promotes *cis* and *trans* target gene transcription by increasing serine-2 phosphorylation of RNA polymerase II (Pol II-Ser2p). We defined an HIV-1 TAR RNA-like (TAR-L) motif in PSA eRNA that was required for CYCLIN T1 binding. Using TALEN-mediated gene editing we further demonstrated that this motif was essential for increased Pol II-Ser2p occupancy levels and CRPC cell growth.

Conclusions: We identify a previously uncharacterized function of eRNA and a P-TEFb activation mechanism and reveal aberrant eRNA expression as a surrogate of abnormality of AR functions in CRPC that may serve as a potential therapeutic target.

Conflict of Interest: The authors declare no conflict of interest.

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