

## **Circular RNAs are differentially expressed in prostate cancer and are potentially associated with resistance to enzalutamide**

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**BACKGROUND:** Most forms of castration-resistant prostate cancer (CRPC) are dependent on the androgen receptor (AR) for survival. While, enzalutamide provides a substantial survival benefit, it is not curative and many patients develop resistance to therapy. Although not yet fully understood, resistance can develop through a number of mechanisms, such as AR copy number gain, the generation of splice variants such as AR-V7 and mutations within the ligand binding domain (LBD) of the AR. Circular RNAs (circRNAs) are a novel type of non-coding RNA, which can regulate the function of miRNA, and may play a key role in the development of drug resistance. circRNAs are highly resistant to degradation, are detectable in plasma and, therefore may serve a role as clinical biomarkers.

**METHODS:** AR-V7 expression was assessed in an isogenic model of enzalutamide resistance. The model consisted of age matched control cells and two sub-lines displaying varied resistance to enzalutamide. circRNA profiling was performed on the panel using a high throughput microarray assay. Bioinformatic analysis identified a number of differentially expressed circRNAs and predicted five miRNA binding sites for each circRNA. miRNAs were stratified based on known associations with prostate cancer, and targets were validated using qPCR. **RESULTS:** Overall, circRNAs were more often down regulated in resistant cell lines compared with control (588 *vs.* 278). Of particular interest was hsa\_circ\_0004870, which was down-regulated in enzalutamide resistant cells ( $p \leq 0.05$ , *vs.* sensitive cells), decreased in AR positive cells ( $p \leq 0.01$ , *vs.* AR negative), and in malignant cells ( $p \leq 0.01$ , *vs.* benign). The associated parental gene was identified as *RBM39*, a member of the U2AF65 family of proteins. Both genes were down-regulated in resistant cells ( $p < 0.05$ , *vs.* sensitive cells).

**CONCLUSION:** This is one of the first studies to profile and demonstrate discrete circRNA expression patterns in an enzalutamide resistant cell line model of prostate cancer. Our data suggests that hsa\_circ\_0004870, through *RBM39*, may play a critical role in the development of enzalutamide resistance in CRPC.

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