

Characterizing PRC1 as a Mediator of Enzalutamide Resistance in Advanced Prostate Cancer

Noah Younger MD PhD,^{*1} Derek Xiang BS,¹ Xiaolin Zhu MD PhD,¹ Shu Chen PhD,¹ Donna Dang PhD,² Lisa Chesner PhD,³ Lisa Kim PhD,² Tony Hua PhD,² Meng Zhang PhD,² Arian Lundberg PhD,² Haolong Li PhD,² Jim Melnyk PhD,² Raunak Shrestha PhD,² Jun Zhu MD,² Emily Egusa,² Rahul Aggarwal MD,¹ Alan Ashworth PhD,⁴ David Quigley PhD,³ Felix Feng MD.²

* = Presenting Author

University of California San Francisco Depts of Medicine¹, Radiation Oncology², Urology³ and HDFCCC⁴

Background

Androgen receptor pathway inhibitors (ARPIs) such as enzalutamide bind directly to the androgen receptor (AR) to antagonize androgen signaling and robustly increase patient survival in advanced prostate cancer. However, not all patients respond to treatment, and resistance inevitably develops with prolonged exposure through mechanisms which are poorly understood. We therefore sought to identify mediators of response and resistance to enzalutamide.

Methods

We used an *in vitro* genome-wide CRISPRi proliferation screen in LNCaP cells +/- enzalutamide treatment to identify a set of genes mediating resistance to enzalutamide. We validated findings using organoid and mouse xenograft models of prostate cancer, followed by mechanistic studies including RNA-seq and CUT&RUN. Finally, we correlated clinical outcomes in localized prostate cancer with differential PRC1 expression levels.

Results

Our CRISPRi screen identified multiple putative enzalutamide resistance genes. Several among the top hits are components of the Polycomb Repressive Complex 1 (PRC1), an epigenetic regulatory complex which represses target gene expression through several mechanisms. We have validated these findings in prostate cancer cell line, organoid, and xenograft models. Targeting PRC1 with small molecule inhibitors synergizes with enzalutamide to slow proliferation. Further investigation has demonstrated increased androgen signaling activity and a decreased neuroendocrine lineage signature in prostate cancer cells with inactivated PRC1, as well as co-localization of PRC1 and AR, suggesting a role for PRC1 in directly modulating androgen signaling and promoting a de-differentiated cell state. Analysis of patient cohorts demonstrates elevated PRC1 expression in persistent/recurrent prostate cancer following hormonal therapy.

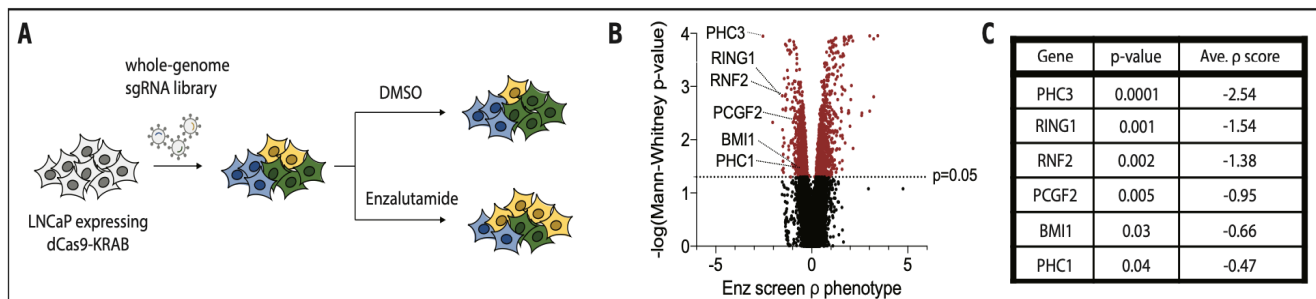


Figure 1: Whole-genome CRISPRi screen for modulators of response to enzalutamide. **(A)** Screen schematic. **(B)** Volcano plot of screen results. Red dots represent statistically significant hits. $p < 0$ = decreased proliferation when combined with enzalutamide. **(C)** Hits that are components of cPRC1.

Conclusions

We have performed an *in vitro* whole genome CRISPRi screen which nominates PRC1 as a driver of enzalutamide resistance in prostate cancer and have validated this result in multiple models. We find evidence in patient cohorts that PRC1 expression correlates with poor response to hormonal therapy. Further, small molecule inhibitors of PRC1 sensitize prostate cancer to treatment with enzalutamide. Our mechanistic studies show that PRC1 modulates transcriptional activity and AR signaling in particular to promote a more de-differentiated, less AR-dependent cell state.

Overall, we have characterized the previously unappreciated role of PRC1 in promoting resistance to enzalutamide, validated therapeutic targeting of PRC1 *in vitro* as a novel treatment approach in prostate cancer, and begun to explore the use of PRC1 expression as a biomarker of response to hormonal therapy. Our hope is that this work will directly benefit patients with advanced prostate cancer by pioneering a new therapeutic strategy as well as by developing a new predictive biomarker of treatment response to hormonal therapy.

Funding Acknowledgments

Research reported in this abstract is supported by the Prostate Cancer Foundation (YIA to NY), Department of War (CDMRP PCRP PRA to NY), and American Society of Clinical Oncology (YIA to NY).

Conflicts of Interest Disclosure Statement

The authors declare no conflicts of interest.