Epigenetic reprogramming with antisense oligonucleotides enhances the effectiveness of androgen receptor inhibition in castration-resistant prostate cancer

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Background
Advanced prostate cancer initially responds to androgen deprivation therapy (ADT); however inevitably recurs as castration-resistant prostate cancer (CRPC). At first sensitive to abiraterone and enzalutamide, CRPC invariably becomes non-responsive to these agents. Novel approaches are required to circumvent CRPC resistance pathways. Our group previously showed the histone lysine-N-methyltransferase EZH2 is overexpressed in prostate cancer and quantitatively associated with progression and poor prognosis. While EZH2 has been described as a transcriptional repressor that methylates histone H3 lysine 27 (H3K27Me3) to mediate epigenetic silencing of multiple tumor suppressors, studies also suggest that EZH2 may regulate androgen signaling in neuroendocrine prostate cancer (NEPC). Nonetheless, whether inhibition of EZH2 can impact AR signaling during resistance to current AR targeting therapies outside of NEPC has not been defined.

Methods and Results
In this study, we screened a library of epigenetic inhibitors for their ability to render CRPC cells sensitive to enzalutamide. We found that EZH2 inhibitors specifically potentiated enzalutamide-mediated inhibition of proliferation. Moreover, we defined antisense oligonucleotides (ASO) as a novel drug strategy to ablate EZH2 and AR expression, which may have advantageous properties in certain settings. RNA-seq, ChIP-seq, and ATAC-seq demonstrated that EZH2 inhibition to alter the AR cistrome, significantly upregulating AR signaling. This suggests an enhanced dependence of CRPC cells on this pathway following inhibition of EZH2. Combination treatment with ASO targeting EZH2 and AR transcripts inhibited prostate cancer cell growth *in vitro* and *in vivo* better than single agents.

Conclusion
In sum, this study identifies EZH2 as a critical epigenetic regulator of ADT resistance and defines ASO-based co-targeting of EZH2 and AR as a promising strategy for treatment of CRPC

Conflict of Interest Statement
YK and ARM are employees of Ionis Pharmaceuticals, which developed the ASOs against EZH2 and AR that were used in this study

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