Epitranscriptomic determinants of enzalutamide resistance

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Background: The METTL3 RNA methyltransferase and RNA N6-methyladenosine (RNA-m6A) modification plays important roles in gene expression, splicing, and translation. RNA-m6A is the most common epitranscriptomic mRNA modification[1] and is dynamically regulated by the METTL3 RNA-methyltransferase complex[2], and FTO[3], ALKBH5 RNA-demethylases[4, 5]. The precise position where METTL3 deposits RNA-m6A within mRNA during transcription is encoded by the epigenetic context of the locus[6]. Thus epigenetic information is positionally and functionally embedded in the epitranscriptome and thereby influences gene expression. We[7, 8] and others[9] have implicated METTL3 in PCa. New therapies targeting METTL3 are in early stage clinical trial for haematological malignancies and solid tumours and could be repurposed for PCa.

Methods: Expression of METTL3 was assessed in a cohort of US prostate cancer patients using immunohistochemistry and clinical-pathologic correlations assessed. Enzalutamide-resistance (EnzR) cell lines were developed, and transcriptomic analysis (RNA-seq) completed between parental and EnzR cells. Methylated RNA immunoprecipitation sequencing (MeRIPSeq) was used to map the transcriptome-wide distribution of m6A. The effect of long and short term enzalutamide treatment on METTL3 was assessed using western blotting.

Results: We have previously shown that METTL3-knockdown or inhibition regulated AR expression, splicing, and androgen signalling in PCa. METTL3 expression is elevated in PCa as compared to adjacent non-malignant PCa. The transcriptome-wide distribution of m6A differs in enzalutamide-sensitive and enzalutamide-resistant PCa cells. Short and long-term enzalutamide treatment alters the expression of AR RNA-m6A regulators in PCa cell lines. Long-term enzalutamide treatment significantly and distinctly altered the transcriptome of PCa cell lines. Further work will determine the potential benefit of RNA-m6A regulator inhibitors for PCa patients.

Conclusions: Targeting METTL3 and RNA-m6A represents a novel approach to fine-tune AR expression and androgen signalling in PCa that could prevent or bypass resistance to existing ADTs.

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