BITS :: Call for Abstracts 2024 - Oral communication

Type	Oral communication
Session	High Resolution RNA Computational Biology
Title	METTL3 inhibition modulates RNA structure, A-to-I editing and immune response in leukemia models
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Affiliation

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Motivation

Among internal post-transcriptional modifications of mRNA, N6-methyladenosine (m6A) is the most prevalent. This modification impacts key processes, including hematopoiesis, and it is catalyzed by the writer enzyme METTL3 (Lan et al., Cancer Res. 2021). Studies have reported METTL3 overexpression in hematopoietic malignancies, among which acute myeloid leukemia (Halene, Tebaldi & Viero, Hematology, 8th ed 2022). We have recently found that conditional deletion of METTL3 in murine fetal liver activates an aberrant innate immune response mediated by the formation of endogenous double-stranded RNAs (dsRNAs) and hematopoietic failure (Gao, Vasic, Song et al., Immunity 2020). Furthermore, the adenosine deaminase enzyme family ADAR is known to catalyze A-to-I editing in dsRNAs to disrupt their conformation (Nishikura, Annual Rev Biochem. 2010). For these reasons, we decided to study the effects of pharmacological METTL3 inhibition on RNA structure, A-to-I editing and immune response, with the goal of modulating dsRNA formation as immunotherapy in acute myeloid leukemia.

Methods

This study comprises a multi-omics analysis of data obtained from human and murine leukemia cell lines treated for 48 hours with METTL3 inhibitors developed by STORM Therapeutics (Guirguis et al., Cancer Discov 2023). RNA-seq was employed to observe the overall effects of METTL3 inhibition on gene expression and splicing. This was coupled with dsRNA immunoprecipitation and sequencing (dsRIP-seq), a recent technique for the detection of dsRNA molecules (Gao et al., STAR Protocols 2021). Using an ad-hoc computational pipeline we were able to identify aberrant dsRNAs that form upon METTL3 inhibition. Changes in A-to-I editing sites were quantified from RNA-seq data with an adapted pipeline from Bullseye (Flamand et al., Genes & Development 2022). Data regarding known m6A sites was retrieved from databases m6A-Atlas v2.0 (Liang et al., Nucleic Acid Res. 2024) and RMBase v3.0 (Xuan et al., Nucleic Acid Res. 2024). Finally, the integration of these data was essential to obtain a comprehensive overview of the effects of METTL3 inhibition in leukemia models.

Results

Here, we show that pharmacological inhibition of METTL3 induces an innate antiviral immune response in human and murine leukemia cell lines. This is characterized by strong activation of the OAS sensors, normally tasked with the detection of foreign dsRNAs, and by the accumulation of aberrant endogenous dsRNAs. Although dsRNAs are specific to each of the systems studied, they share common properties: they present long 3’ UTRs, they are highly m6A modified in their native state and they are characterized by low folding energies. Additionally, METTL3 inhibition results in the upregulation of the cytoplasmic isoform of the adenosine deaminase enzyme ADAR1 and in an increase in A-to-I editing events, specifically in the 3’ UTR regions of m6A modified mRNAs. These data point to A-to-I editing as a possible rescue mechanism for the stimulation of dsRNA-mediated immune response in METTL3 inhibited cells.
Collectively, these results suggest that combined inhibition of METTL3 and ADAR1 can be used to modulate dsRNA formation as a potential immunotherapy against leukemia and other tumors characterized by high METTL3 activity.

Info

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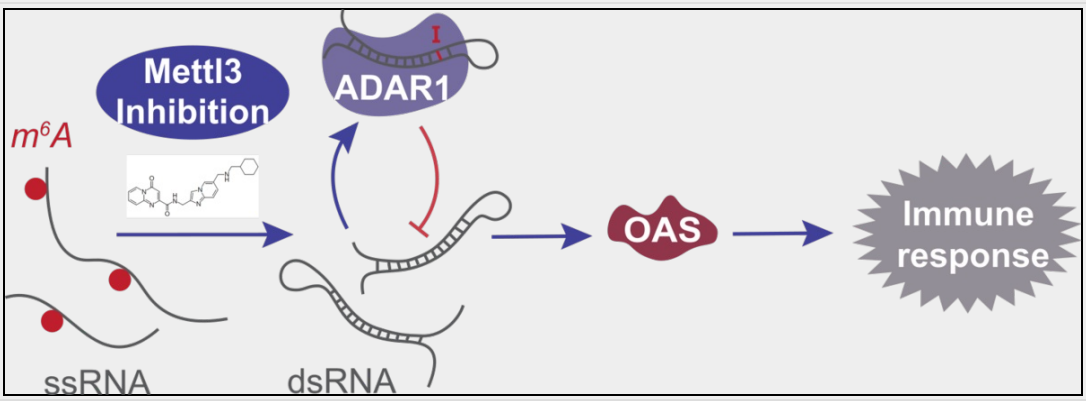
Figure



Availability	https://docs.google.com/document/d/1VlvUYNq5Z_-6xC_8BzmYhrQ8RFeFJpsyBKKCLfqj_XI/edit?usp=sharing

Dissemination Material

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Summary

Study of the effects of pharmacological METTL3 inhibition on RNA structure, A-to-I editing and immune response, with the goal of modulating dsRNA formation as immunotherapy in acute myeloid leukemia.

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Submitted on	02.05.2024