

RMEL3 lncRNA Drives Melanoma Cell Survival and Immune Modulation via Isoform-Specific Functions

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Background: Our group previously identified RMEL3 as a long non-coding RNA (lncRNA) enriched in BRAFV600E-mutant melanoma and proposed a positive feedback loop involving RMEL3/MAPK/PI3K pathways. Despite its functional relevance, the structural features and molecular mechanisms of RMEL3 remain unclear. Here, we expanded the analysis of structure, function and molecular interaction of RMEL3 in melanoma cells. **Methods:** We used 5′/3′ RACE, nested RT-PCR, RNA-FISH, the MS2/MCP-GFP system, doxycycline-inducible exon-specific shRNAs, chemically modified exon-specific antisense oligonucleotides (ASOs), RNA pulldown with mass spectrometry and Western blotting, protein immunoprecipitation followed by RT-qPCR, DNA sequencing, and RNA-seq. **Results:** We identified the major transcription start site (TSS) of canonical RMEL3 and found it enriched in open chromatin marks specifically in melanoma. One circular and several novel linear RMEL3 isoforms were identified, including one driven by an intronic single nucleotide polymorphism. RMEL3 is enriched in transposable elements and transcript diversity results from alternative TSS, splicing, splice junction usage, polyadenylation, and Alu element-mediated termination. Endogenous RMEL3 localized mainly to cytoplasmic particles/condensates, while MS2-tagged isoforms localized to the cell cortex or condensates depending on the variant. RMEL3 interacted with MAPK and HIPPO pathway components and *in silico* analysis identified conserved stem-loop structures predicted to mediate RNA–protein interactions. Knockdown reduced proliferation, migration, and invasion, increased cell death, and decreased phosphorylated ERK and AKT levels upon serum re-stimulation. RNA-seq revealed differential expression of genes involved in signaling, the cell cycle, actin cytoskeleton, extracellular vesicle biogenesis, and notably innate and adaptive immune responses. IFN-γ stimulation reduced canonical—but not circular—RMEL3 expression, confirmed by RT-qPCR and RNA-seq studies. Isoform-specific ASOs effectively silenced RMEL3, with differing anti-proliferative effects, and reproduced immunoregulatory gene changes. **Conclusion:** Ongoing studies aim to clarify isoform-specific effects on proliferation and whether IFN-γ-mediated RMEL3 repression contributes to apoptosis, cell cycle arrest, or resistance to IFN-γ and immunotherapies in melanoma.

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