

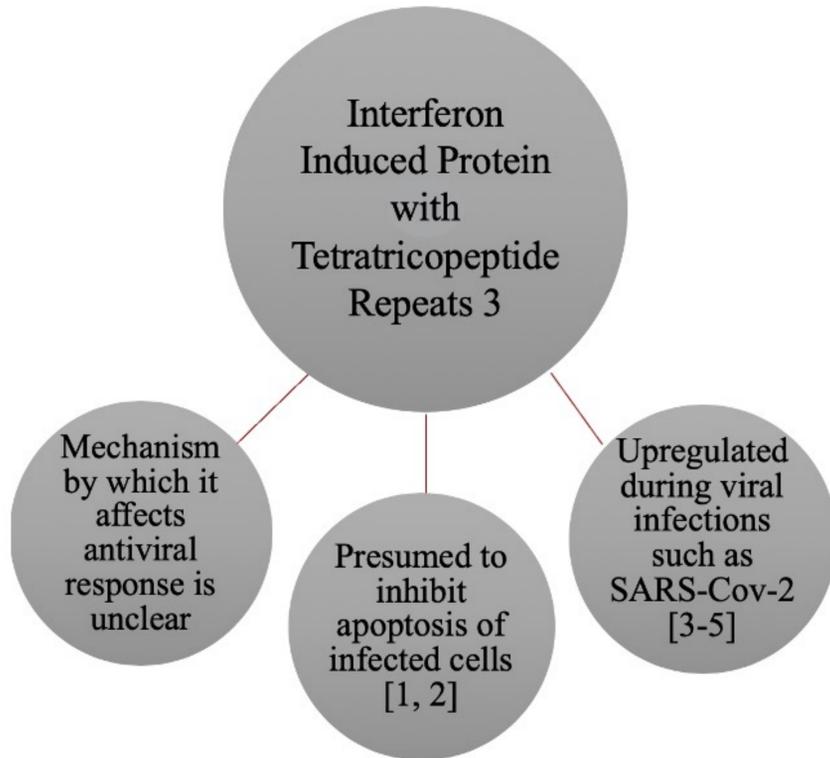
Characterization of the Anti-Apoptotic Effects of IFIT3 During the Antiviral Innate Immune Response in an IFIT3 Knock-Out Cell Line



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Introduction



Objectives

- Characterize the mutations of IFIT3 in IFIT3 Knock-Out respiratory epithelial cell lines
- Determination of the effects of IFIT3 on apoptosis during the innate antiviral immune response

Figure 1. Plasmid treated with CRISPR-Cas9 to target IFIT3 for mutation. A549 cells were transfected with pSpCas9-IFIT3 plasmid to generate cell lines that knocked out IFIT3.

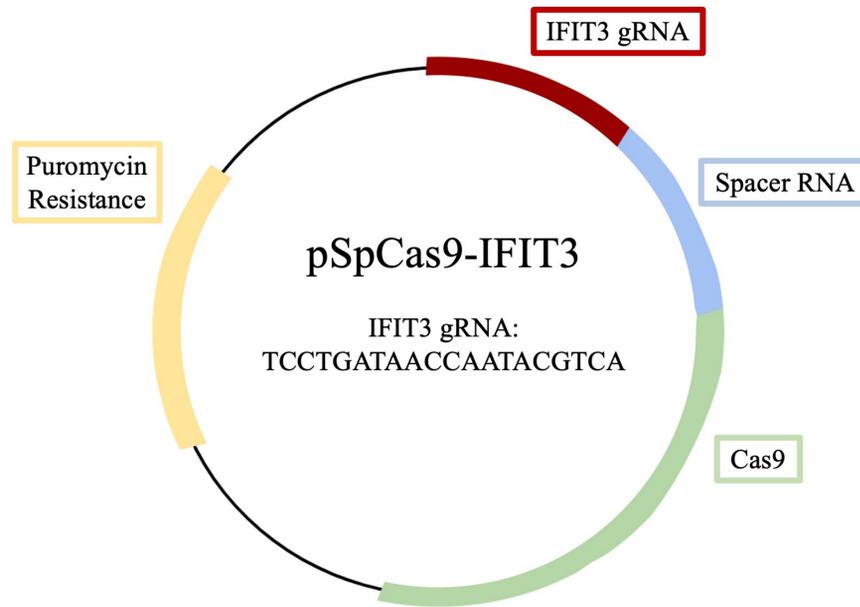
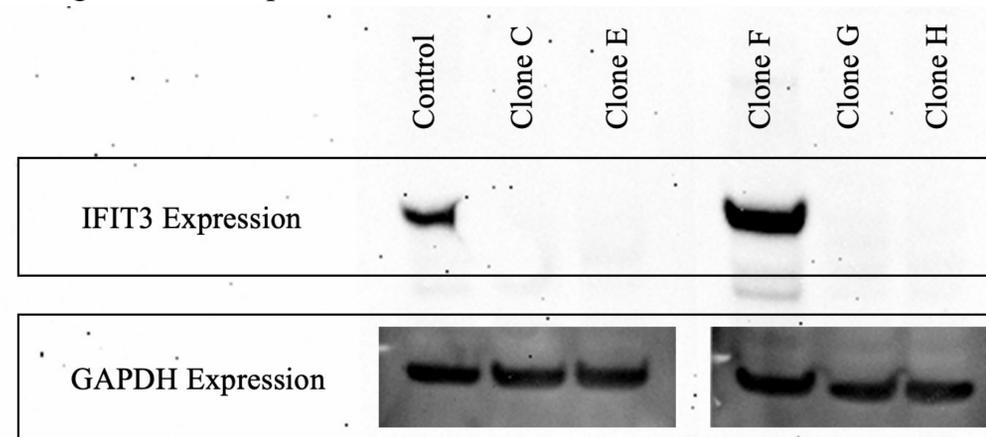


Figure 2. Confirmation of Clones C, E, G, and H as IFIT3KO Cell Lines. Suspected IFIT3KO cell lines (as well as control A549 cells) were transfected with Poly I:C for 16 hours and evaluated for potential knockouts via Western Blotting. GAPDH expression is also shown.



Methodology

- Treated A549 cells with CRISPR-Cas9 plasmid to knock out expression of IFIT3
- IFIT3KO cells treated with genomic PCR so that mutations could be sequenced

Future Directions

- IFIT3KO cell lines treated with Poly I:C and monitored over 72 hours to quantify differences in activation of apoptotic pathways
- Greater presence of Caspase-3 protein in IFIT3KO cells
- More IFIT2 aggregation in IFIT3KO cells

Acknowledgements

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