Tumor Suppressor microRNA-34a Inhibits Glioblastoma Cell Proliferation and Sensitizes to Temozolomide and Radiation

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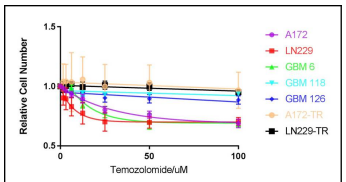
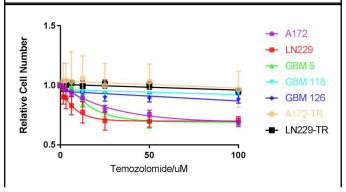
Introduction

Different combinations of three driver mutations localized respectively to the p53, Rb and receptor tyrosine kinase networks are responsible for selective growth advantage of glioblastoma tumors (GBM) and contribute to significant intra-tumoral heterogeneity, a major cause of treatment failure. microRNA-34a (miR-34a) modulates the expression of multiple genes in all three deregulated GBM networks, suggesting it could serve as a novel therapeutic agent.

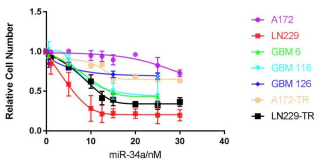
Methods & Results



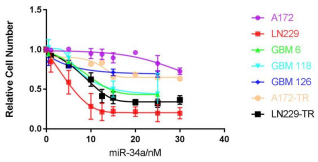
GBM cell lines and primary cultures are variably sensitive to Temozolomide and cells with acquired resistance can be generated from parental cell lines



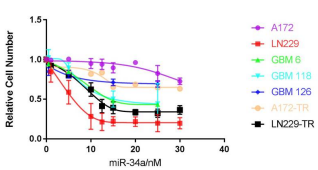
miR-34a inhibits proliferation in all GBM cell lines and primary cultures



Cells were reverse transfected with miR-34a mimics or control microRNA in 96 well plates and relative cell number was determined through sulforhodamine B assay

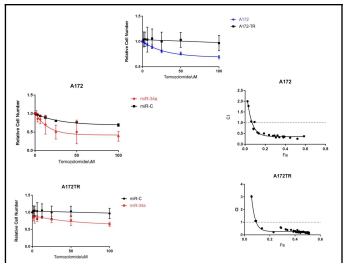


Transfection with miR-34a mimics results in significant inhibition in proliferation of all GBM cell lines and cultures

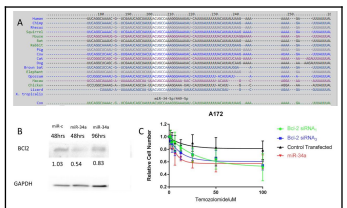


Transfection with miR-34a mimics significantly reduces proliferation in all GBM cell lines

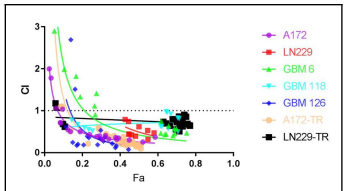
miR-34a sensitizes to Temozolomide in multiple tested cell lines irrespective of the baseline temozolomide sensitivity



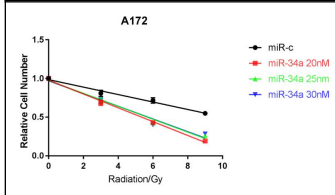
a172-a172 TR curves



A: The 3'UTR of Bcl2 mRNA has a broadly conserved site for the 5' seed region of miR-34a. B: Transfection with miR-34a mimics results in significant decrease in protein expression of Bcl2. C: Sensitization approximating that of miR-34a can be reproduced by two different siRNA targetting Bcl2

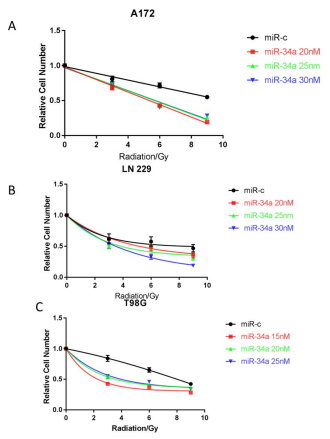


miR-34a sensitizes to Temozolomide therapy



miR-34a sensitizes to radiation

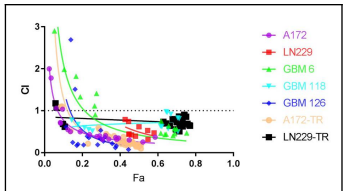
miR-34a sensitizes to Radiation in multiple GBM cell lines



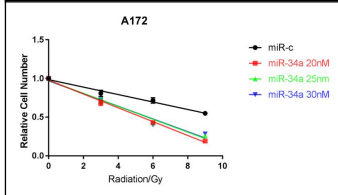
A,B and C: Transfection with miR-34a mimics sensitizes to Radiation in all tested GBM cell lines with Dose Enhancement Ratios of 1.7-2.2. Cells were reverse transfected with miR-34a mimics and treated with 3,6 and 9 Gy of Radiation in 96 well plates 48 hours post transfection.

Conclusions

miR-34a is a promising novel therapeutic that inhibits proliferation and sensitizes to TMZ and radiation. Current work is focused on delivering miR-34a to orthotopic tumors using different types of nanoparticles.



miR-34a sensitizes to Temozolomide therapy



miR-34a sensitizes to radiation