MR-Guided Focused Ultrasound Delivery of Polymeric Brain-Penetrating Nanoparticle MicroRNA Conjugates in Glioblastoma

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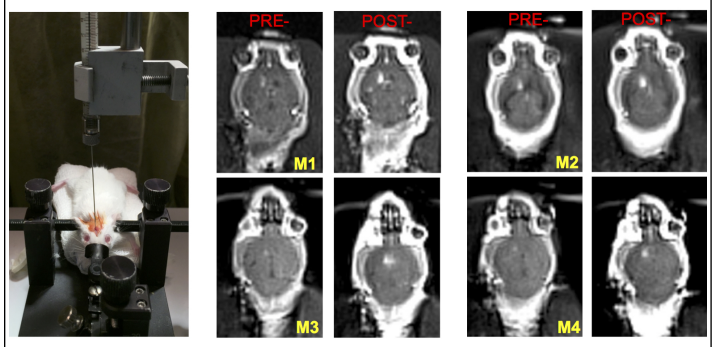
Introduction

MicroRNAs (miRNAs) are small noncoding RNAs that regulate gene expression by targeting the mRNAs of a large number of human genes. We have previously demonstrated that miRNAs could serve as therapeutic agents for Glioblastoma (GBM). However, systemically administered genes to the CNS is hindered by both the blood-brain barrier (BBB) and the nanoporous electrostatically charged tissue space, denoted here as the "brain tissue barrier" (BTB). In order to overcome both of these physical barriers we have designed a non-invasive targeted gene delivery approach. We use nanoparticle (NP) delivery, as they offer the potential for enhanced transfection efficiencies and controlled-drug release.

Methods

To deliver gene-bearing NPs across the BBB, we use focused ultrasound (FUS) and contrast agent microbubbles (MBs). FUS was applied using MR-guidance. We have shown that activating MBs with FUS yields safe and transient BBB opening in the FUS focal zone. Technologies for overcoming the BTB center on coating the miRNA bearing NPs with an extremely dense brush layer of polyethylene glycol (PEG). NPs are injected at the time of BBB opening to permit their delivery to the CNS.

MR-Guided FUS for BBB Disruption

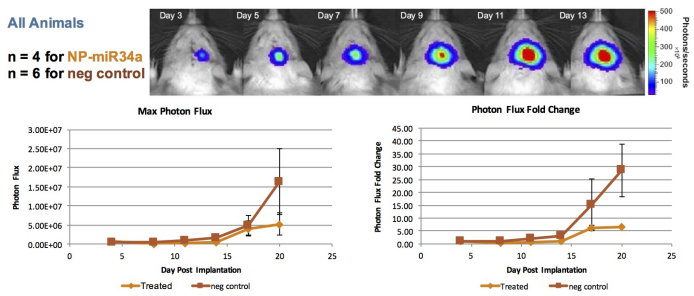


(Left) Stereotactic implantation of U87-GBM cells. (Middle/Right) Representative treatment group demonstrating the disruption of the BBB. MRI imaging reveals increase contrast uptake before and after MR-guided FUS.

Results

We used a blend of non-PEGylated and highly PEGylated polymers at an optimized ratio to engineer brain-penetrating DNA NPs with a poly-ethylenemine (PEI) core polymer. We delivered PEI-NPs (~60 nm) carrying either a control scramble plasmid or miRNA-34a (tumor suppressive, GFP or luciferase) plasmid DNA across the BBB in mice using MR-guided FUS-MBs. Robust luciferase transgene expression, corresponding to a single focal site of FUS exposure, was visible, and the intensity of gene expression was correlated with PEI-NP concentration. After delivering miR34a PEI-NPs across the BBB with FUS-MBs, we immunochemically detected GFP in both glial cells and neuronal cell nuclei. miR34a expression was homogeneously distributed throughout the sonicated area, demonstrating the benefit of combining FUS-mediated delivery across the BBB with brain penetrating NPs.

MR-Guided FUS Treatment of miR-34a PEI Nanoparticles



(Above) Representative bioluminescence IVIS scans acquired after treatment. (Below) Non-invasive in vivo monitoring of U87-GBM tumor growth via luciferase. Results suggest that there is increased cell death with the PEI-NP miR-34a subgroup when compared to the scramble miR-control.

Learning Objectives

By the conclusion of this session, participants should be able to: 1) Describe the use of MR-guided focused ultrasound in GBM; 2) Discuss the role of miRNAs in regulating pathways in GBM; 3) Identify an effective treatment using both MR-guided ultrasound and nanoparticle therapies.

Conclusions

Our results indicated that we can use MR-guided FUS to deliver miRNAs across the BBB as a treatment modality in GBM. Going forward, this approach will be used for the concerted regulation of gene expression by miRNAs and their effects on GBM malignancy.

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