Determing glioblastoma proteome changes in response to lateral ventricle neural stem cells

Lauren Whaley¹, Emily S Norton², Nataanial Zarco², Hugo Guerrero-Cazareza

¹ University of North Florida, Jacksonville FL, 32224; ² Department of Neurosurgery, Mayo Clinic, Jacksonville, FL, 32034, USA

Background

Glioblastoma

- Glioblastoma (GBM) is the most common and malignant primary tumor in adults.
- GBM tumors located near the lateral ventricle display a more aggressive recurrence pattern, negatively impacting patient survival.

- Suggests involvement of subventricular zone neurogenic niche in GBM malignancy.

Methionyl-tRNA synthetase (MetRS)

- Mutant MetRS L274G (MetRS*) allows for incorporation of azido-labeled methionine analog azidonorleucine (ANL) into newly formed proteins.

- MetRS* metabolic labeling can be successfully channeled into a lentivirus and utilized as a tool for cell-specific proteomics with the use of ANL.

- GBM cells within close proximity of neural stem cells show an increase of proteins representative of malignant cancer spread.

Methods

- Molecular cloning of MetRS* into lentiviral backbone with puromycin selection
- Confirmation of ANL incorporation into multiple MetRS mutant cell lines using western blot and silver stain
- Confirmation of ANL incorporation into multiple MetRS mutant cell lines using western blot and silver stain

- Titration
- Lentivirus production

- Puromycin selection of transduced cells

Conclusions

- Methods for co-culturing with neural stem cells
- Proteomic changes in GBM cells

References

- Link, A. James; Vink, Mandy K. S.; Tirrell, David A. Journal of the American Chemical Society (2004), 126(4), 1050-10602. DOI:10.1021/ja038472e