Transfecting B35 Neuroblastoma Cells with Near Infrared Protein for Photoacoustic Detection

Christina Aridi, Biomedical Engineering
Mentor: Barbara Smith, Ph.D.
School of Biological Health Systems Engineering

Research Question

The overarching goal of this research is to isolate a monoclonal cell line expressing iRFP to calibrate a custom PAM system for depths beyond current optical based microscopes.

Overview

Viewing biological processes at depth in tissues is essential for the study and diagnosis of neurodegenerative and neurological diseases. Despite recent advancements, current optical techniques are restricted to an imaging depth of ~1mm due to the scattering of light in tissues [1]. In combination with near-infrared fluorescent proteins (iRFP), photoacoustic microscopy (PAM) systems can use light and sound to image specific cell types deeper in biological tissues than traditional optical microscopy methods. Our custom PAM system in combination with iRFP enables effective non-invasive methods to actively detect and analyze specific neurons at increased depths.

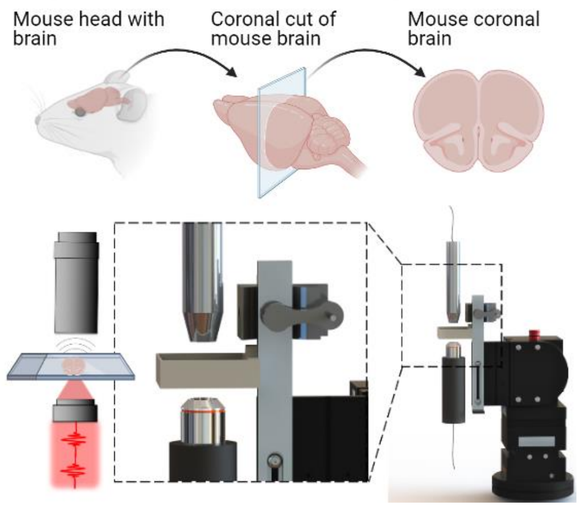

Fig 1: PAM system imaging a mouse brain slice through a glass slide.

Experimental Design

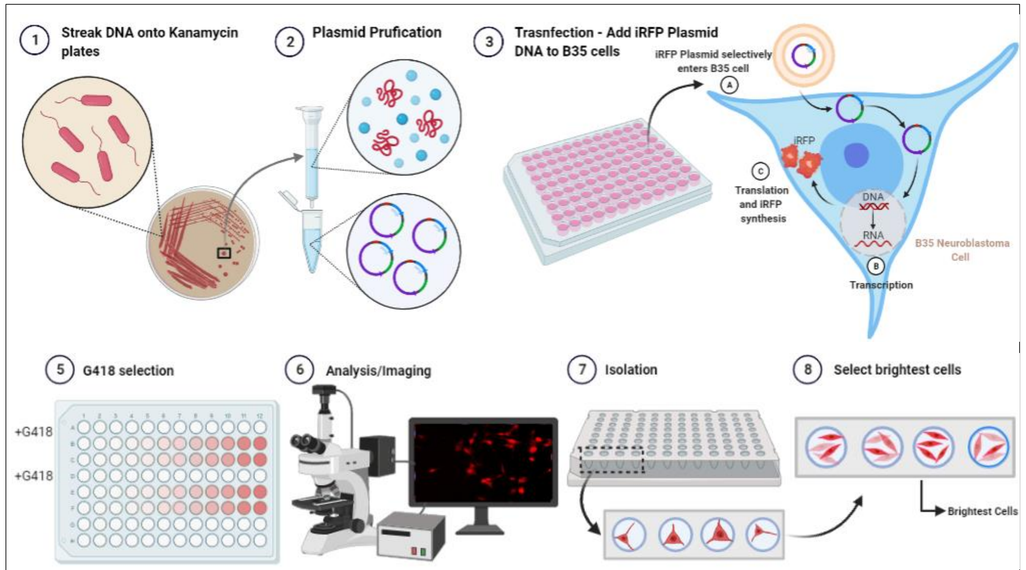

Fig 2: Schematic showing the experimental design process.

Results

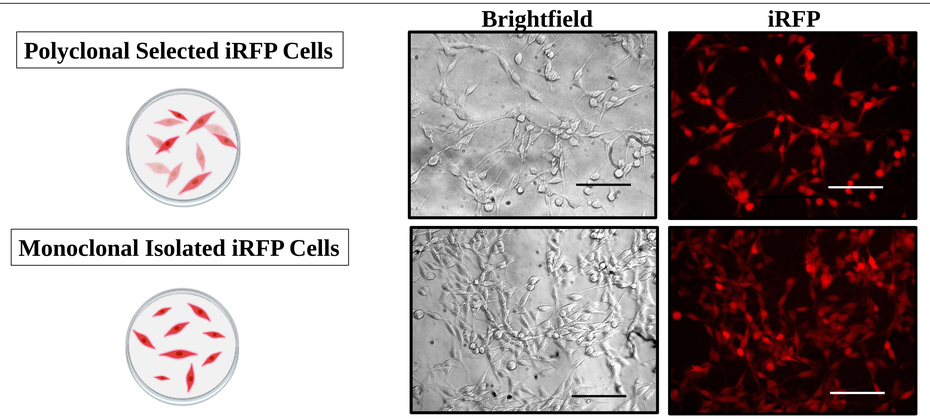

Fig 4: Fluorescence microscopy images of transfected polyclonal B35 neuroblastoma cells (top). Fluorescence images of monoclonal B35 neuroblastoma cells (bottom). Scale bar is 100 microns.

Conclusion

This research demonstrates the ability to successfully transfect B35 neuroblastoma cells with iRFP. In this research, we purified plasmid DNA, transfected B35 cells with Lipofectamine and the bacterial plasmid, and stably transfected iRFP-expressing monoclonal lines, as pictured in Figure 4.

Plasmid DNA Characterization

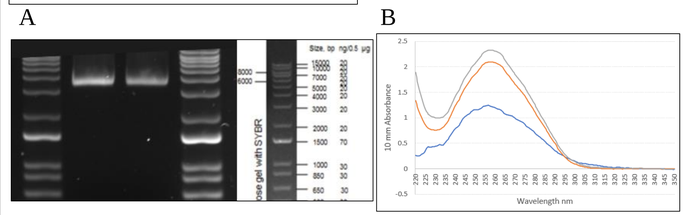

Fig 3: A) The gel electrophoresis verifies that the plasmid is present, since it reached between 4000-5000 base pairs. B) Nanodrop data was used to determine the quantity and quality of the plasmid.

Future Work

The transfected B35 neuroblastoma cells will be used to calibrate a custom PAM system for detection of specific cell types in tissue.

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References

[1] Wang, Lihong V, and Song Hu. “Photoacoustic tomography: *in vivo* imaging from organelles to organs.” *Science (New York, N.Y.)* vol. 335,6075 (2012): 1458-62.

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Ira A. Fulton Schools of Engineering
Arizona State University