SWIFTY FOUNDATION

YOUR 2019 CO-FUNDING IMPACT REPORT

Alex's Lemonade Stand

WITH GRATITUDE

Dear Ginny, Patti and Al,

We want to sincerely thank you for your support, and even more so, for your commitment to helping find better treatments and cures for medulloblastoma. Because of your incredible efforts, preliminary data has been developed, and exciting research has been published. Because of you and your commitment to fighting childhood cancer, we are closer to a day where there will be cures for all kids with this heinous disease.

Thank you for your continued support – we are truly honored to have the Swifty Foundation by our side in the fight against childhood cancer. Please let me know if there is anything you need from us here at ALSF!

Until there's a cure,

Melanie Gould
Alex's Lemonade Stand Foundation

Targeting Eya2 to Inhibit c-Myc driven Medulloblastoma Tumor Progression

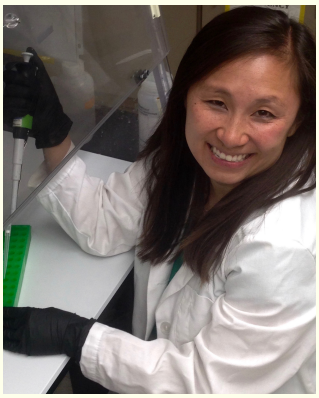
Dr. Melanie Vincent, University of Colorado, Denver

2016 Young Investigator Grant

Project Timeline: July 1, 2016 to June 30, 2019

"Thanks to the funds provided by The Swifty Foundation, we have promising data that shows targeting Eya2, a developmental protein that is inappropriately turned on in Group3 Medulloblastoma patients, can inhibit c-Myc driven tumor growth in Group3 Medulloblastoma."

Project Update



Medulloblastoma is a deadly pediatric brain cancer. Although there are different types of medulloblastoma, patients are uniformly treated with therapies including surgery, radiation and chemotherapy.

To identify novel therapeutics for children with c-Myc driven medulloblastoma, we investigated the role of the developmental protein Eya2, which can partner with another developmental protein Six1 or act by itself, to control c-Myc levels. Our preliminary data showed that targeting Eya2 provides new means to inhibit c-Myc expression in these tumors, which is the driving force of this cancer.

We are collaborating with investigators at A*STAR to develop Eya2 small molecule inhibitors with better drug-like qualities so that we can test them in animal models. We are excited to target Eya2 with our inhibitors because Eya2 is an excellent therapeutic target for these patients since Eya2 is only turned on in the tumor and not in normal healthy tissues. Thus, they may provide a novel therapeutic option that avoids the debilitating side effects that occur from currently available treatments.

Thanks to the funds provided by The Swifty Foundation, we have promising data that shows targeting Eya2, a developmental protein that is inappropriately turned on in Group3 Medulloblastoma patients, can inhibit c-Myc driven tumor growth in Group3 Medulloblastoma. Our genetic proof of principle experiments helped support further investigation of our small molecules that inhibit Eya2. We hope that our preliminary data will drive the continued research of our small molecule inhibitors to hopefully lead to a possible targeted therapy option for these children. We are very excited about the data and truly grateful for the support from the Swifty Foundation and Alex's Lemonade Stand Foundation!

Updates on Dr. Vincent's project will be available after receipt and review of her final report in August 2019.

Pre-clinical Testing of Novel Glutamine Metabolic Inhibitors in MYC-driven Medulloblastoma

Dr. Eric Raabe, The Johns Hopkins University School of Medicine

2016 Innovation Grant

Project Timeline: September 1, 2016 to August 31, 2018

Project Update

We have improved drugs that can block key metabolic pathways downstream of cancer causing genes, and we are working to find other drugs that can be used in combination to help patients who are in need of new approaches to cure their cancer.

We have identified two such combinations and are working to move these into further testing. Funding from ALSF and the Swifty Foundation has helped us generate the preliminary studies that have allowed us to obtain long-term funding from the National Institute of Health.

We have just learned that the first manuscript of this Swifty Foundation/ALSF-funded project was accepted for publication. Entitled *"Orally bioavailable glutamine antagonist prodrug JHU-083 penetrates mouse brain and suppresses the growth of MYC-driven medulloblastoma,"* this manuscript describes the first-generation new metabolic inhibitor as an effective treatment for aggressive medulloblastoma. We show that the inhibitor kills medulloblastoma cells in the dish and extends the life of mice with two different strains of high-MYC medulloblastoma.

We are working hard on a second manuscript which identifies another metabolic drug that can cooperate with our glutamine metabolic inhibitor

to enhance the killing of high-MYC medulloblastoma. These publications will support the further advance-ment of DON and DON prodrugs into a pediatric phase I clinical trial.

Future Plans

One part of the next phase of this research is to continue to develop new prodrugs, building on what ALSF and the Swifty Foundation have helped us to discover so far.

We are going to test the ability of these glutamine-altering prodrugs to alter the immune microenvironment in immune-intact mice bearing syngeneic models of human MYC-driven medulloblastoma. We are working to further develop these combinations with DON prodrugs, and we anticipate that our study will culminate in a clinical trial.

"We have just learned that the first manuscript of this Swifty Foundation/ALSF-funded project was accepted for publication... These publications will support the further advance-ment of DON and DON prodrugs into a pediatric phase I clinical trial."

De-escalation of Radiotherapy for Medulloblastoma by a Novel DNA Damage Checkpoint Inhibitor

Dr. Marc Symons, The Feinstein Institute for Medical Research
2017 Innovation Grant
Project Timeline: January 1, 2018 to January 1, 2020

Support from the Swifty Foundation is supporting Dr. Symons as he tests a novel approach to reducing the harmful side effects of radiation.

Project Update

Our studies aim at reducing the side effects of radiation therapy in children with medulloblastoma, a brain tumor that can metastasize through the spine.

Over the past year, we have implemented and optimized a genetic model of the most aggressive form of this disease. We are now examining whether minocycline, an anti-inflammatory drug, can sensitize medulloblastoma tumors to radiation, which should allow us to treat children with lower doses of radiation, thereby protecting their brains. Minocycline already is in clinical use as a neuro-protective drug, and we therefore expect that this would further diminish the side effects caused by radiation.

In addition, our recent results showed that minocycline also strongly diminishes spreading of medulloblastoma cells into surrounding normal brain, and we therefore expect to see that minocycline also will prevent or diminish medulloblastoma metastasis.

Future Plans

This year, we will test both minocycline and simvastatin for their potential to radiosensitize medulloblastoma. For the studies, we need to identify a dose of radiation that will provide an equivalent increase in survival as the combination of a lower dose or radiation and the drug (minocycline/simvastatin).

Once we identify such dose, we will proceed with the behavioral experiments that will compare radiation alone with the combination of radiation and both TAM-inhibiting drugs. If time permits, we also will perform a detailed analysis of the effects of minocycline and simvastatin on Myc/DNp53 medulloblastoma metastasis to the spine.

Exploring synthetic lethality of one-carbon metabolism genes and mTORC1 inhibition in MYCN amplified Medulloblastoma

Shannon Wong-Michalak, University of California San Francisco
2018 POST Grant
Project Timeline: May 21, 2018 to July 16, 2018

Final Report Summary

During my time as a POST Scholar, I gained invaluable experience performing research on medulloblastoma, the most common solid tumor in children. In addition to contributing data to a larger project, I developed my skills as a researcher through learning new techniques, designing experiments, and learning how to troubleshoot assays when they don't work out ask planned.

For my part of the project, I was generating CRISPRi viruses that would knock down gene candidates from a ribosome profiling screen. The screen identified genes that were translationally, but not transcriptionally, down-regulated in response to an mTOR inhibitor that slows the growth of the tumor cells. We wanted to test whether any of these genes were responsible for the decreased tumorigenicity of the cells observed in response to the drug.

First, I went through the gene list to see whether any of the genes on the list had been previously implicated in other cancers. The one-carbon metabolism pathway is extremely important for cancer cells because among the products are purines, required for DNA synthesis, and methylation reactions, which regulate epigenetic expression. I wanted to see whether these genes would effect the growth of medulloblastoma cells.

Next, I generated viruses that would incorporate the gRNA specific to these genes into the medulloblastoma lines containing the dCAS9 protein. I wanted to see whether knocking down this gene would decrease the rate of proliferation. Preliminarily, I did see a decrease in proliferation relative to my negative control. However, the experimental conditions would need to be optimized before I would be confident in saying that it had a true effect. Additionally, I generated gRNA viruses for a number of other genes that my mentor will use to investigate other candidates.

In addition to the new lab techniques and greater understanding of medulloblastoma I learned, this experience taught me about the patience, diligence and determination required to be a scientist.

"My summer in with the POST program has inspired me to continue my involvement in pediatric cancer research. This year, I am continuing to work at the Weiss Lab at UCSF as a lab technician, assisting a postdoctoral scholar on her project concerning neuroblastoma. I am also applying to medical school. With my medical degree, I hope to become a pediatric oncologist and treat patients with the cancers that I have spent my time researching in the Weiss Lab."

Publications & Presentations

Below is a sampling of publications and presentations resulting from ALSF/Swifty Foundation funding, self-reported by researchers in their final reports and follow-up correspondence.

Allison Hanaford, Brad Poore, Jesse Alt, Barbara Slusher, Charles Eberhart and Eric Raabe. Abstract 3484: In vivo metabolomics reveals a potentially potent combination therapy for MYC-driven medulloblastoma. AACR Annual Meeting 2018; April 14-18, 2018; Chicago, IL DOI: 10.1158/1538-7445.AM2018-3484. Published July 2018.

Allison Hanaford, Charles Eberhart, Eric Raabe. Novel oral prodrugs of 6-diazo-5-oxo norleucine improve brain penetration and demonstrate efficacy against myc-driven orthotopic medulloblastoma xenografts. Neuro-Oncology, Volume 19, Issue suppl_4, 1 June 2017, Pages IV 59, Pediatric Basic and Translational Research Conference New York City, June 2017.

Eric Raabe, et al. Orally bioavailable glutamine antagonist prodrug JHU-083 penetrates mouse brain and suppresses the growth of MYC-driven medulloblastoma. Accepted for publication, 2019.

Melanie Vincent, et al. Identifying the individual roles of eya2 in myc-driven group 3 medulloblastoma. Abstract for Poster Presentation. CU Anschutz Medical Campus, T32 Pediatric Hemotology and Oncology Symposium. Aspen, CO, September 2017.

Melanie Vincent, et al. “Targeting Eya2 in Myc-driven Group3 Medulloblastoma.” Abstract for Poster Presentation. CU Anschutz Medical Campus, Pharmacology Retreat. Keystone, CO, April 2018.

Micah Maxwell, Brad Poore, Allison Hanaford, Jesse Alt, Rana Rais, Barbara Slusher, Charles Eberhart and Eric Raabe. Abstract 3521: Glutamine metabolic inhibition synergizes with L-asparaginase in MYCN amplified neuroblastoma. AACR Annual Meeting 2018; April 14-18, 2018; Chicago, IL. DOI: 10.1158/1538-7445.AM2018-3521 Published July 2018.