UNIVERSITY OF LOUISVILLE
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Division of Hematology and Oncology
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raiseRED
2024-2025 Impact Report
2025-2026 Research Initiatives

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2022-2024 Report on Research Fund Utilization

Introduction

We are grateful to raiseRED for your generous support of pediatric hematology, oncology, and cellular therapy research. Your funding has supported novel work conducted by Dr. William Tse, Dr. Michael Huang, Dr. Megan Scruggs, and Dr. Kayla LaRosa. Unfortunately, Dr. Huang departed for another institution in January 2025 and sadly, Dr. Tse unexpectedly passed away in March of 2025 which left a gaping hole in their research programs. Dr. Michael Ferguson and Dr. Jun Cai have taken up the mantle to lead these projects particularly on the groundbreaking Chimeric Antigen Receptor T-cell (CAR-T) work that Dr. Tse had started. Through Dr. Ferguson and Dr. Cai's work, the projects were able to get back on on schedule as the work is highlighted below, but did leave the amount budgeted to be less than expected which will be rectified with the next academic year's spending. Again, we are forever grateful to raiseRED for the generous support and the projects below show our dedication to innovative research that can change treatment paradigms for children with cancer.

Dr. Tse/Ferguson/Cai – CAR-T

Dr. Tse's CAR-T cells have been developed to combat a particularly deadly type of brain tumor called Diffuse Intrinsic Pontine Glioma (DIPG). The median survival rate for children with DIPG is 11 months and is almost universally fatal at 2 years. Chemotherapy and radiation only delay the disease progression and work into novel therapies have been greatly needed. Initial work in CAR-T cell therapies have shown some minor success in clinical trials, but issues with CAR-T cell exhaustion and inhibition from the surrounding tumor cells, termed the "tumor microenvironment" has limited this success As stated in the previous report, Dr. Tse's research group has pioneered several innovative strategies utilizing a novel CRISPR-Cas9 gene editing technique to introduce the new CAR receptor into the normal (endogenous) T cell receptor. Additionally, Dr. Tse has introduced in his CAR-T cell an IL-2Rβ receptor with a STAT3/5 binding motif that increases signaling in the STAT3/5 pathway. The combination of these two methods has reduced exhaustion of these CAR-T cells and enhanced the effectiveness of their tumor cell killing.

Dr. Cai's group in collaboration with Dr. Tse, developed a xeno-transplant mouse model of DIPG, meaning human DIPG cells were injected into the brainstem of immune-deficient mice which reliably grew similar to human disease. Utilizing Dr. Tse's CAR-T cells (termed CRISPR-STAT3/5-CAR-T), with just one infusion, over 75% of the mice showed complete tumor disappearance. We have now monitored the mice for over a year with no tumor recurrence detected. Additional testing in mice with a humanized immune system are underway to see if the innate immune system will alter the efficacy of these CAR-T cells. Due to this innovative work, Dr. Tse was awarded the Oncology Plenary Session/Talk at the American Society of Pediatric Hematology Oncology (ASPHO) Annual Meeting in May of 2025 at which Dr. Ferguson was able to present his work. Drs. Cai and Ferguson submitted a full patent application for this

technology after Drs. Cai, Tse, and Ferguson submitted a provisional patent application last year. Drs. Cai and Ferguson are working additional mouse toxicity studies in hopes of presenting their work for an Investigational New Drug (IND) meeting with the FDA in 2026 so that they move this CAR-T cell to clinical trials.

Dr. Huang/Ferguson – Immune Microenvironment of High Grade Brain Tumors, DFMO treatment for Medulloblastoma

Dr. Huang's research prior to his departure delved into characterizing the immune cell landscape in high-grade and low-grade brain tumors in order to understand the immunosuppressive properties of this tumor immune microenvironment. Understanding the interference from the immune system will help us better tailor treatments to be more effective and potentially less toxic. Dr. Ferguson was able to take over this work, given his background in genomics, to look at immune cells in tumor tissue and matching genetic changes that indicate immune involvement and dysregulation in these tumors. In conjunction with Dr. Muge Sak from bioinformatics, Dr. Ferguson is in preparation of a manuscript outlining the vast differences in immune cell make up in high-grade versus low-grade tumors that we hope to exploit for treatment modifications.

Additionally, Dr. Huang served as the Study Chair for a multi-institutional protocol investigating the efficacy of DFMO/eflornithine, an anti-parasitic drug, for treating molecular high-risk medulloblastoma—the most prevalent form of childhood brain cancer. With his departure, Dr. Mustafa Barbour is helping lead the way in this pivotal clinical trial run through the Beat Childhood Cancer (BCC) consortium.

Dr. Scruggs – Bone Health in Pediatric Cancer Survivors

Children with solid malignancies receive aggressive multimodal treatments (including chemotherapy, radiation therapy, transplants). As a results of the disease involving the bones and the treatments themselves, these children are at risk for developing poor bone health over time. Studies in long-term childhood cancer survivors have determined that these patients are at risk for low bone mineral density and fractures, poor linear growth, and other musculoskeletal issues. However, no prospective studies currently exist assessing bone health in this patient population during active cancer therapy or shortly after completing treatment. Dr. Scruggs proposed a prospective, multi-institutional study that will assess bone health in children and young adult patients treated at either Norton Children's Hospital or the University of Kentucky Children's Hospital with solid malignancies through the implementation of DEXA scans, laboratory assessments, and a physical therapy assessment throughout their treatment course and shortly after the end of therapy. Dr. Scruggs plans to study if a correlation exists between the bone mineral density values on DEXA scans, physical activity levels, and common laboratory values that describe bone health (vitamin D, calcium, phosphorus, alkaline phosphatase, etc.) and is in the process of getting this study approved and underway.

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Dr. LaRosa – Circadian Rhythm Disruption in ALL

Disruption to the circadian rhythm, the body's internal clock that regulates alertness and sleepiness, may occur because of cancer or its treatment. Youth being treated for cancer may commonly experience what is known as cancer- and treatment-related symptoms (CTRS), including fatigue, sleep difficulties, depressive symptoms, and cognitive impairment. It is hypothesized that cancer treatments, such as surgery, radiation, and chemotherapy, may provoke an inflammatory response within the body and CTRS may occur in reaction to this. Circadian rhythm disruption is hypothesized to be an underlying mechanism of the development of CTRS. Overall, youth with ALL have less stable and robust circadian rhythms, greater fatigue, and were less active when compared to healthy children. Common methods of assessing circadian rhythm disruption include a combination of parent and/or self-report of sleep habits, actigraphy (a wrist-worn device that estimates sleep/wake through movement), monitoring of light exposure and physical activity, and collection of biomarkers of sleep (e.g., saliva, urine). Dr. LaRosa purchased 10 actigraphy watches and light medallions, which will allow for assessment of circadian rhythm disruption. The purchase of these devices would allow for data collection within a small sample of youth receiving treatment for leukemia and this work is currently underway.

2025-2026 Research Initiatives

Given the novel work that is already underway, the hope is to expand our efforts into sarcoma, particularly osteosarcoma, Ewing sarcoma, and rhabdomyosarcoma. All three of these sarcomas when found to be metastatic at diagnosis have cure rates under 30% and we have continued to use the same toxic chemotherapy regimens for the last 30 to 40 years as nothing has been found to be an improvement. Drs. Ferguson and Cai are currently working on developing a mouse model of osteosarcoma based on Dr. Ferguson's previous work at Riley Children's Hospital. They both have worked on additional changes to the CAR-T designed by Dr. Tse to persist in this bone tumor environment that is rich in production of a certain cytokine, TGF-β. This cytokine, TGF-β, has been shown to function as a potent metastasis promoter by fostering tumor growth, inducing transition to malignant cells (i.e. epithelial-mesenchymal transition (EMT)), increasing tumor-associated fibrosis, enhancing new vessel growth (angiogenesis), and more importantly blocking antitumor immune responses in osteosarcoma. Drs. Cai and Ferguson have designed a CAR-T cell that is insentitive to TGF-β effects on T cells and are working to test in the mouse model of osteosarcoma that is being developed.

Also according to last year's report, Dr. Ferguson was in the first steps of investigating a novel epigenetic blood-based biomarker, Oxford Biodynamics EpiSwitch™ technology, to detect disease remanence or recurrence in pediatric solid tumors and brain tumors. Dr. Ferguson has worked with this company to develop a protocol to first study this technology in patients with osteosarcoma who often have small abnormalities on CT scans, particularly in the lungs that often are missed or thought to be infection-related. Our **hypothesis** is that the EpiSwitch® CCS that can be found in peripheral blood can detect recurrence of osteosarcoma earlier, prior to

imaging or clinical changes allowing the oncologist the opportunity to change treatment plan when disease burden is at a minimum. Two of the EpiSwitch blood-based biomarker applications are utilized clinically today in US and UK as reimbursable tests: a Checkpoint Inhibitor Response Test (CiRT) for prediction of treatment response in immuno-oncology, and a prostate cancer screening test (PSE) showing that this technology is useful for diagnostic purposes and response assessment which supports Dr. Ferguson's hypothesis.

Conclusion

Due to the generosity of all the students in the Dance Marathon and raiseRed, we have been able to support transformative research by Drs. Tse, Huang, Scrugss, LaRosa, Ferguson, and Cai. Because of this support, we are on the precipice of bringing some of this work to clinical trials in the next year or two which will offer new treatments and biomarkers for those children with refractory cancers in desperate need. Thank you again for all you do for pediatric cancer and blood disorders. We will continue to work diligently to be good stewards of all your fundraising efforts finding novel treatments for the most recalcitrant diseases.