PROJECT TITLE: Utilizing CRISPR-Cas9 technology to introduce novel variants into AML cell lines

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Project Description

Acute myeloid Leukemia (AML) is the most common leukemia in adults. Molecular features such as cytogenetics and somatic mutations are essential components of risk stratification and are frequently used in daily clinical practice to determine the appropriate treatment modality and intensity (e.g., LeukemiaNet). These advancements in understanding AML biology have led to improved disease outcome, including long-term complete remission (CR) in 40% of patients over 60 years old. However, the establishment of these prognostic factors were based on data generated from large-scale genomic studies of AML patients of primarily Central-European Ancestry. Recent studies by our collaborator Ann-Kathrin Eisfeld’s group (OSUCC) showed that patients of African ancestry (self-identified as Black/African American and confirmed via SNP-array based genotype) exhibited significantly decreased survival compared to individuals of Central-European ancestry (“White”); independent of factors previously affiliated with survival disparity such as socioeconomic status, access to treatment, follow-up, and age. To investigate the underlying source of disparity observed in these patients, we sequenced the exomes of 100 Black AML patients (Alliance) and compared somatic mutation frequencies to those found in 741 White patients (BeatAML). In this analysis, we found 162 recurrently mutated genes, including genes with high variant allele frequency (VAF) that not been previously implicated in AML. One of the most common variants and previously unreported variants observed in this dataset was Pleckstrin-Homology-Domain Interacting Protein (PHIP). We expect the completion of current studies to result in functional data to nominate PHIP as a potent leukemic driver and/or a modifier of treatment response. The goal of this work is to provide an exemplar of heretofore unrecognized mutations in AML and to provide sufficient mechanistic evidence to include PHIP on cancer resequencing panels; changing clinical practice to make risk
assessment guidelines more inclusive for AML patients.

An UPRISE student who joins for our lab for the summer would have the opportunity to learn about molecular oncology research through interactive group meetings and independent experimentation at the research bench. Mentored by postdoctoral researcher Bailee Kain, the student will learn how to introduce knockout mutations in Leukemia cell lines using CRISPR-Cas9 found in our African Ancestry AML dataset. Following confirmation of effective gene knockout, the student will be exposed to multiple research techniques and can receive hands on training in flow cytometry, qPCR, PCR, and western blot. Bailee will also assist the student in learning appropriate data analysis and presentation approaches. Students will be expected to meet regularly with Bailee and Dr. Grimes to discuss their progress, attend journal clubs, and participate in lab meetings.