

The Role of Epigenetic Heterogeneity in CLL Evolution

Dana Farber Cancer Institute

PI: Dan Landau

Grant Number: 1-K01ES025431-01

Chronic lymphocytic leukemia (CLL) is currently incurable. Despite effective treatments, the disease invariably recurs due, in part, to its ability to evolve. We have shown that pretreatment intra-leukemic genetic heterogeneity foreshadows clonal evolution leading to disease relapse. Nevertheless, the cellular phenotype and its fitness for selection result from both genetic and epigenetic alterations. Therefore, a major challenge in the study of cancer evolution is to integrate genetic and epigenetic heterogeneity. In preliminary studies, we found increased intra-sample epigenetic heterogeneity in CLL. To understand the basis of this heterogeneity, we studied the uniformity of the methylation status of neighboring CpGs contained within individual reads from massively parallel bisulfite sequencing of ~100 primary CLL samples. We demonstrated that most of the heterogeneity stems from disordered methylation, a form of stochastic epigenetic drift. Disordered methylation affected regions important to transcriptional regulation and was associated with a decoupling of the relationship between promoter methylation and transcriptional silencing. Finally, disordered methylation was subjected to selection and may facilitate clonal evolution. I hypothesize that disordered methylation impacts histone modification and transcription, thereby enhancing CLL evolution. To define the impact of disordered methylation, we propose the following independent yet interrelated Specific Aims: (1) To examine its relationship to histone modification and transcription, we will produce comprehensive histone ChIP-seq mapping and ChIP-bisulfite-seq directed at repressive histone marks. We will integrate the multidimensional data to infer epigenetic intra-sample heterogeneity and validate this with single-cell RNAseq to assess cell-to-cell variability as a function of methylation disorder. (2) We will develop a statistical inference tool to detect putative methylation "driver" events in cancer taking into account background stochastic variation. (3) To study the impact of disordered methylation on clonal evolution and clinical outcome, we will integrate genetic and epigenetic heterogeneity analysis in pretreatment samples from 350 patients who received uniform treatment, and 80 relapse samples. There are no therapeutic strategies currently available to curb cancer evolution. Thus, these studies address an unmet therapeutic need. Finally, in this application, I have outlined a 5-year career development plan to meet my goal of becoming an independent investigator in translational cancer biology, proficient in big data science methodology. I have assembled a Mentorship Committee of internationally recognized experts to provide scientific and career mentorship. I will pursue intensive didactic coursework and hands-on training with leading experts, to develop a strong computational and statistical foundation. Finally, Dana-Farber Cancer Institute is the ideal environment for attaining my scientific and career goals, given its outstanding research community, emphasis on big data science, and an excellent track record of training independent physician-scientists. PUBLIC HEALTH RELEVANCE: Chronic lymphocytic leukemia frequently undergoes evolution in response to therapy, resulting in a more aggressive and treatment-resistant form of the disease. I propose a new mechanism to explain this evolution, which contributes to greater diversity of subpopulations within the cancer. In this proposal, I will investigate how this mechanism, termed "disordered epigenetic patterning", facilitates leukemia evolution, thereby paving the way for the development of therapeutic approaches to curb the adaptive potential of cancer.