This summer, I rotated in a lab in the department of Hematopoietic Biology and Malignancy with a heavy emphasis on leveraging single-cell genomics techniques to deeply interrogate molecular events underlying the pathophysiology of blood cancers, including acute myeloid leukemia (AML). During my time there, I presented a paper during journal club on a computational tool which utilizes somatic and mitochondrial DNA mutations to infer clonal relationships between cancer cells, especially in AML1. In part of this paper, the researchers utilized known driver mutations to classify cells as healthy, pre-leukemic, or leukemic stem cells. However, I realized that I did not know the pathological and clinical significance of these driver mutations. Therefore, my motivation for this project is to build a solid foundation in understanding 1) what are the common mutations in these genes, 2) the signalling pathways that are implicated / affected downstream, and 3) how these mutations may play a role in prognosis / response to therapy. Of course, as the class progresses and I am introduced to new methods of analyzing biological data, I will consider how to incorporate them into my final report.

The classical view of hematopoiesis holds that hematopoietic stem cells differentiate into two major branches – myeloid and lymphoid. AML, as the name implies, is a blood cancer involving primarily stem cell precursors of the myeloid branch2. Leukemic blasts are characterized by the accumulation of mutations that result in impaired myeloid progenitor differentiation, as well as the rapid proliferation of and evasion of apoptotic signalling. These functional differences allow leukemic blasts to rapidly expand and dominate bone marrow space, interfering with normal hematopoiesis. As such, common initial presenting symptoms are often related to impaired hematopoiesis, such as recurrent infections (low WBC) or bleeding disorders / coagulopathies (platelet aberrancies). To make a definitive diagnosis, a bone marrow biopsy is required.

While the paper I covered in journal club includes a number of known leukemic mutations, I decided to search for a more comprehensive list of mutations to search through and found a review from 2020 covering driver mutations in AML3. This paper lists and categorizes many important functional mutations, including NPM1, considered a “licensing mutation” (pre-leukemic mutation); FLT3-ITD/TKD, KIT, and NRAS/KRAS mutations, which affect signal transduction; RUNX1, CEBPA, and GATA2 mutations, which encode for transcription factors; many epigenetic modifiers, such as DNMT3A, TET2, IDH1/2, ASXL1, EZH2, BCOR/BCORL1, and the cohesin complex; and finally mutations in tumor-suppressor genes such as TP53 and WT1, which are not leukemia-specific.

While there are many interesting driver mutations to study, I decided initially to focus on the three most common mutations in AML: NPM1, FLT3, and DNMT3A. After further consideration, however, I have decided to ultimately narrow my focus to just NPM1 and FLT3. These two mutations appear to have some interplay in that assigning a risk category for a patient’s AML is dependent on the states of both genes4. For instance, mutated NPM1 without FLT3 (or low levels of mutated FLT3) falls in a favorable risk category. Mutated NPM1 and high FLT3-ITD is considered intermediated risk, but so is wild-type NPM1 without FLT3-ITD or with FLT3-low. In addition, wild-type NPM1 with FLT3-ITD high is considered adverse risk. Therefore, it appears that it is the **mutated** form of NPM1 that confers a lower risk category. If possible, I would like to spend part of this project to take a closer look at how these two driver mutations are related (and if this effect is limited to a particular mutation). The other benefit of choosing these two mutations is that they are the most common mutations seen in AML, which means that there will be high-quality information with well-annotated metadata in public databases readily available for analysis. For instance, just a cursory search in dbSNP for NPM1 shows 12789 results, and 37972 results for FLT3. In dbVar, there are 144 results for NPM1 and 506 for FLT3. Therefore, I believe that there will be a good wealth of data to explore and draw conclusions from.

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