Multidisciplinary Research Program in Medicine Project*: High-throughput cell isolation for cancer genomics using inkjet printing*

Hypothesis or Research Question(s): A novel device that incorporates optimized inkjet dispensing and machine vision can enable rapid and accurate isolation of single cells for genomic and transcriptomic high-throughput sequencing applications.

PROJECT BACKGROUND & SUMMARY

The cell is the basic unit of life, and new methods for profiling the genetic characteristics of individual cells are now transforming our understanding of early development, tissue regeneration, immunology and disease. Cancer arises when a single cell acquires genetic alterations leading to uncontrolled replication. As tumour cells divide they continue to acquire mutations which they pass on to their descendants, forming sub-populations with different characteristics. This genetic diversity can allow tumours to evolve resistance to treatment and eventually spread. As such, profiling the genetic alterations and patterns of gene expression within tumours and their surrounding environment can reveal mechanisms underlying cancer relapse and metastatic progression.

Existing single cell omics platforms present deficiencies in throughput, accuracy and customizability. We are developing an inkjet-based instrument that integrates high-resolution imaging and neural network-based object recognition to rapidly isolate cells for genetic profiling. Building on a working prototype recently installed in the Genome Sciences Centre at the BC Cancer Research Institute, we will engineer new features to ensure accurate cell classification, temperature control, and ease of operation. We will validate the platform by implementing protocols for single cell DNA and RNA sequencing, benchmarking efficiency and data quality relative to an existing commercially available instrument.

The project requires multidisciplinary expertise spanning biomedical and electrical engineering, molecular biology, and bioinformatics. Key steps in a single cell genomics workflow include: cell culture or tissue dissociation, single cell isolation, preparation of nucleic acid libraries, high-throughput sequencing, and analysis of large-scale datasets. Feedback from each step in the process will be used to optimize earlier steps. As such, collaboration and cross-disciplinary communication will be critical for the success of the project.

Single cell genomic technologies are being widely adopted in academic research, as well as in the biotechnology, pharmaceutical and diagnostic sectors. The device under development will provide an open and customizable platform for a range of single cell omic workflows, permitting in-depth characterization of cellular composition in healthy tissue and disease.

BENEFIT TO THE STUDENTS

The project involves a collaboration between Principal Investigators from the Departments of Medical Genetics/BC Cancer and Electrical Engineering.

An undergraduate student with a background in engineering will assist with implementing new platform features. The prototype device is an inkjet spotter capable of dispensing single cells onto a substrate in preparation for sequencing. The undergraduate will have the opportunity to work alongside an engineering PhD student to support implementation of new components, which will involve assisting with programming a front-end user interface for device operation, and helping with design and implementation of a temperature controlled substrate to preserve cell viability. Applications such as RNA sequencing will require rapid, direct dispensing of cells into lysis buffer or onto a chilled substrate. TS1 will integrate a Peltier plate cooler, cooling block, temperature controller, sensor, and power supply

to deliver a temperature-controlled substrate with operational range of 4-70˚C and stability ±0.25˚C stability over 1 hour. Through this project, TS1 will gain experience in instrumentation and hardware design.

A student with experience in computer programming (background in biology is an asset) will contribute to bioinformatic analysis of high-throughput single cell sequencing data derived from the instrument. TS2 will have the opportunity to gain experience with the Linux command line and Python workflows, as well as parallelization in a high performance computing (HPC) cluster environment. They will contribute to genomic data processing, quality control and visualization, summarizing key features of large-scale datasets and feeding back information to their engineering collaborators, TS1 and PhD1, for further optimization.

A medical student, if available, will contribute to biological sample processing. The device should be tested with a range of cell lines and tissue samples in order to ensure efficient and accurate performance. TS3 will use previously established protocols to digest solid primary tissue into a single cell suspension. Under optical imaging, cellular debris can manifest as small irregularly shaped remnants of dead cells, or larger undigested clumps of cells and extracellular matrix; the presence of debris can confound downstream bioinformatic analysis. TS3 will also use previously established protocols to isolate cell nuclei. TS3 will work with TS1 to use the instrument to acquire datasets of these preparations, which will be labeled to enable neural network training for expanded classification. TS3 will gain experience with cell culture and tissue digestion techniques in order to prepare high-quality, viable single cell suspensions for sequencing.

All students will have the opportunity to learn about the research process in a dynamic and collaborative interdisciplinary environment. By the end of the summer they will prepare a presentation on their contribution and progress, and will have an opportunity to contribute to a research manuscript. This experience will help TS to develop interdisciplinary communication skills, while fostering critical thinking through planning and troubleshooting their experiments. These practical skills (including programming, engineering design and validation, and tissue culture) will be valuable to their future careers, in industry, medicine, or in preparation for graduate studies.