REQUEST FOR PROPOSALS
Single-cell RNA-seq Pilot and Feasibility Awards

The Chao Family Comprehensive Cancer Center (CFCCC) is pleased to announce that funding is available to support use of single-cell RNA-sequencing (scRNA-seq) for cancer research through the UCI Genomic High-Throughput Facility (GHTF) Single-Cell Analysis Core.

BACKGROUND

DNA and RNA profiling has provided some of the most important insights into the complex etiology and diversity of tumors. These insights have been magnified by the ability to probe at single-cell resolution. The GHTF Single-Cell Analysis Core supports Cancer Center researchers by providing access to single-cell gene expression technology, analysis, training and integration. The 10x Chromium single cell 3’ gene expression technology can analyze up to 10,000 single cells. Single cells are encapsulated with reagents and a Gel Bead containing barcoded oligonucleotides in nanoliter-sized vesicles called Gel Beads in Emulsion, or GEMS. Within the emulsion droplet, lysis of the cell and dissolution of the Gel Bead allows barcoded reverse transcription of polyadenylated mRNA. Library construction for the Illumina platform continues with the cDNA in a single bulk reaction. The library is sequenced on a lane of the HiSeq 4000 and the “Cell Ranger” analysis pipeline performs sample demultiplexing, barcode processing (deconvoluting to single cells) and single cell 3’ gene counting. The total cost for library preparation and sequencing, including all consumables, reagents to generate the single cell libraries, quality control assays and Illumina sequencing, is $3,800.

In order to recover the expected number of cells, it is important to maximize viability and minimize the cell preparation time. The ideal situation is a suspension of more than 90% viable cells with the cell concentration measured using a hemocytometer or similar cell counter. During preparation of the single cells suspension, it is best to use wide-bore pipette tips and gentle centrifugation conditions (150rcf – 300rcf). The cells should be washed in 1xPBS containing 0.04% weight/volume BSA to minimize cell losses and aggregation. It is recommended that the cell suspension be filtered using an appropriate strainer, e.g. MACS SmartFilter. The optimal cell concentration is ~1,000 cells/µL and, if targeting to capture 10,000 cells, the total number of cells used for encapsulation will be 17,400.

PURPOSE

The goals of this funding opportunity are twofold: (1) to support investigators’ use of scRNA-seq through the GHTF to acquire preliminary data for external cancer-related grant applications; and (2) to encourage the use of scRNA-seq in their research by investigators who are new to the technology.

FUNDING

The Cancer Center will provide a maximum of $3,000 per project, which will be augmented by support from 10x Genomics to cover one sample for single cell library preparation, sequencing on one lane and a Cell Ranger Report. Ten projects will be awarded on a competitive basis.

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Funds will be transferred directly to the GHTF to be spent, as specified, to subsidize usage fees for the awarded projects. **Funds must be spent by March 15, 2019.**

**ELIGIBILITY**

Any UCI faculty investigator is eligible to apply. The proposed scRNA-seq experiment should be designed to produce preliminary data supporting an extramural peer-reviewed cancer-related grant application, or to test the feasibility of scRNA-seq as an approach to a cancer-related research question. If the PI is not a current CFCCC member, the faculty member would be expected to apply for CFCCC membership as a condition of award.

**PROPOSAL REQUIREMENTS**

Using the application form provided, applicants should submit the Principal Investigator’s NIH biosketch and a completed application form that includes an abstract, lay summary, and a brief (one page max) project description that details the sequencing project and discusses how it furthers an ongoing project or improves the likelihood of acquiring external funding for a new project. Cancer Center Senior Leadership will evaluate the proposals and notify awardees when decisions have been made. Only one application will be accepted per investigator. Potential applicants are strongly encouraged to discuss projects with GHTF staff prior to submission.

**APPLICATION DEADLINE**

Please submit the completed application as a single pdf to bmcclelnd@uci.edu by 5:00 PM Friday, September 14, 2018.

**CONTACTS**

General questions related to the RFP or the Cancer Center should be directed to the Cancer Center Program Office at 714-456-3628 or bmcclelnd@uci.edu.

Questions related to scRNA-seq and the GHTF should be directed to Dr. Melanie Oakes, Genomic High-Throughput Facility Manager, at 949-824-6023 or ucightf@gmail.com.