

Dear Genomics and Microbiome Core Facility (GMCF) investigators:

The GMCF team would like to thank you for your confidence in us to handle your precious samples! We have just completed our fiscal year (FY25) and are now well into our new fiscal year (FY26) and would like to report on some changes to our workflows and new capabilities. As you will see, we have added new instruments and capabilities, but we are saying goodbye to some instruments too! See below for a list of changes to our capabilities. We look forward to continuing to support your research efforts going forward. Please reach out to any of us with questions/comments/requests. Just a reminder that project requests can be submitted through Bookitlab (<https://core.bookitlab.com/rush-GMCF/Login>). Please also look for our updated sample and data retention policy, which includes storage of all samples for 3 months after completion unless you ask for longer and archiving of Illumina data in Basespace after six months. PacBio data will be available for download for 7 days after completion and then archived. Data retrieval from archive will, unfortunately, incur additional charges. Please reach out to our Rush Research Bioinformatics Core (RRBC; rrbc@rush.edu) for help with data analysis, data storage/transfer, submission to the NCBI Sequence Read Archive (SRA), and more.

Best wishes from the GMCF!

Changes include:

- (1) We have **acquired** a new Illumina DNA sequencer – the **Illumina MiSeq i100+**. This instrument is meant to replace the old MiSeq instrument, and has many advantages, including: (1) 4X faster runtime, (2) higher quality, particularly for 2x300 sequencing runs, (3) room-temperature reagents (no dry ice!), (4) greater tolerance for amplicons, (5) lower cost per sequence, and (6) greater flexibility in flow cell size. The i100+ instrument currently has two flow cell sizes of 5M and 25M clusters, but additional flow cells of 50M and 100M clusters are expected by the end of the year. Current read lengths are 2x150 or 2x300, but Illumina is developing 2x500 chemistry for this instrument that is expected to be released in October (25M clusters). One major difference between the i100+ instrument and the prior MiSeq is the use of patterned flow cells. This increases the robustness of the instrument, but also makes the instrument susceptible to index hopping. Therefore, only unique dual index (UDI) libraries should be sequenced on the instrument. Libraries generated from combinatorial indices (CDI) may lead to improper assignment of sequence data to barcodes. We suggest avoiding CDIs.
- (2) We are **discontinuing** service for the old MiSeq (including Nano, V2, and V3 flow cells). Due to the discontinuation of the old MiSeq and MiniSeq (see below), we will no longer be processing samples with Fluidigm CS linkers. All incoming amplicons should have unique dual indexes (UDI). Please reach out to us to get sequences of the linkers for 1st stage amplicon library preparation that are compatible with UDI barcoded adapters.

- (3) We will be **discontinuing** service for the Illumina MiniSeq, but the instrument will be available for use until it breaks. Libraries with combinatorial indices (CDIs), rather than UDIs, can be sequenced on the MiniSeq. Maximum output for mid-output flow cells is 8M clusters (2x150).
- (4) We are **discontinuing** service for the NovaSeq6000 instrument (including SP 2x250 flow cells). We have options to replace this (see below); please reach out to us for help in choosing the best platform for your work.
- a. Illumina MiSeq i100+ with 2x300 and possibly 2x500 sequencing for 5M, 25M or 50M cluster outputs.
 - b. Illumina NovaSeqX with 2x300 on the 1.5B (billion) cluster flow cell (2 lanes of 750 M clusters each)
 - c. Complete Genomics G-800 instrument with 1x600 base sequencing on a 1.6B cluster flow cell (4 lanes of 400 M clusters each)
- (5) We have **acquired** a new DNA sequencer from **Complete Genomics – the G-800** (<https://www.completegenomics.com/products/sequencing-platforms/dnbseq-g800/>). This sequencer has expected quality scores in the Q40 range (rather than Q30) and has individual reads of 600 bases (currently single direction, 1x600). Further increases in read-length are expected. Complete Genomics suggests improved coverage in difficult-to-sequence genomic regions. Standard Illumina libraries can be converted (by GMCF) to be compatible with Complete Genomics. Each instrument can run two flow cells independently. Available flow cell sizes include:
- a. 1x100 – 1.8B clusters
 - b. 2x100 – 1.8B clusters
 - c. 2x150 – 1.8B clusters
 - d. 1x600 – 1.6B clusters
- (6) We have **acquired** a new liquid-handling robot from **Volta Labs** called the **“Callisto Sample Prep System”** (<https://www.voltalabs.com/product>). While this instrument can make PacBio, Oxford Nanopore, Illumina and Complete Genomics libraries, we specifically acquired this instrument to help with hybridization capture protocols. Currently the instrument can perform IDT capture protocols, with Twist captures coming soon. We will be happy to conduct a wide range of off-the-shelf and custom capture protocols, including: whole exome sequencing (WES), whole transcriptome capture – particularly effective for damaged (e.g., FFPE) human DNA, viral captures, etc. Please reach out to us for any projects of interest.
- (7) We have **acquired** two **Singulator 200 devices** from **S2 genomics** (<https://s2genomics.com/singulator-platform/>). This instrument is a “...flexible, automated

solution for the rapid dissociation of solid tissue into high-quality, viable single cells or intact nuclei compatible with numerous single-cell genomics applications.” As a reminder, we have two platforms for single-cell RNAseq library preparation, including the 10X Chromium X device and associated chemistries, as well as the Illumina Single Cell Prep (formerly Fluent). For spatial transcriptomics, we operate the 10x Visium platform with a CytAssist device and work closely with the UIC spatial core facility for projects needing the 10x Xenium platform.

- (8) We are currently running **full-length 16S ribosomal RNA gene amplicon sequencing** protocols using the **PacBio Kinnex** chemistry (<https://www.pacb.com/technology/kinnex/>). We have developed a custom, adaptable two-stage PCR protocol that can be deployed for amplicons in the range of 700-2000 bp. Turn-around time is slower than with Illumina sequencing, but data are very high quality (Q30-Q40) and total output from a single SMRT cell on a PacBio Revio instrument is on the scale of 40M reads. The GMCF can provide you with portions of a run (minimum 10%), just as we do for Illumina sequencing.
- (9) We will be adding a new **charge for development of new primer sets**. Adapting new primer sets is labor intensive and requires the use of one or more agarose gels. Thus, we will be charging one-time flat fees of \$50 per new primer set when temperature optimization is necessary and \$25 per new primer set when investigators provide us with established annealing temperatures.

Contact Information

Laboratory	Name	Email	Title	Focus areas
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