



Gene Expression Center

University of Wisconsin-Madison Biotechnology Center

Parse scRNA-Seq Service Policy

We highly recommend mock measurements be performed on extra samples of all cell types to ensure satisfactory sample preparation quality prior to fixing your experimental samples. An automated fluorescence cell counter is available in the GEC.

If cells are to be sorted, it is the client's responsibility to coordinate and schedule sorting with UWCCC Flow Core.

Client is responsible for assessing cell viability and aggregation of each sample prior to fixation according to the protocol being followed. We recommend you document (with images, if possible) the condition of the cells at the time of fixation. We cannot guarantee a successful experiment based on these cell counts alone and strongly encourage you to plan a small pilot experiment (e.g., with the 10,000 cell Mini Kit) first if your goal is a much larger submission.

Samples submitted for service will be counted and assessed on an automated cell counter with acridine orange and propidium iodide (AO/PI) viability stain 1-3 days before start of service. Cell counts will be compared to submitted cell numbers and if there is >20% variability client will be contacted.

Every effort will be made to provide a successful and satisfactory experience but unfortunately, we cannot guarantee the results. Within reason, we will do our best to assist with troubleshooting if needed on a per project basis at the discretion of the core director.

Guidelines

- Before designing your experiment or starting sample fixation, we recommend referring to the Evercode Fixation user manual and contacting us at gecinfo@biotech.wisc.edu to arrange a meeting with GEC staff and Parse technical support representatives.
- The recommended sample input for fixation is a minimum of 100,000 cells or nuclei and a maximum of 4 million cells or nuclei. As many cells or nuclei as possible should be fixed to avoid recovering fewer cells than desired downstream. If feasible, start with >1 million cells or nuclei.
- Parse recommends a hemocytometer for cell counting but alternative devices can also be used. The GEC uses the viability stain AO/PI to assess sample quality.
- Before fixation, cell suspension should be >70% viable and <5% cell aggregation/debris as recommended by Parse. When quantifying fixed samples, it is critical to avoid counting cell debris to avoid overestimating the number of cells/nuclei.
- Please prepare a 12ul cell counting aliquot of each sample following fixation. These aliquots will be thawed and counted a day or two before proceeding with the Parse protocol to account for any changes during the storage and freeze thaw.
- Ensure the 1.5 mL and 15 mL centrifuge tubes are polypropylene, as polystyrene tubes will lead to substantial sample loss.
- Fixed samples should be transported/shipped on dry ice to avoid any possible freeze/thaw of the sample.

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