**SORTING EXPERIMENT PROTOCOL**

*This form must be filled out and received by the CMtO Facility at the start of the project or at least a* ***week*** *before the actual Sort appointment*

Name: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ Email:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

Date of Experiment: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_ **Phone Number:\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_**

Laboratory/Principal Investigator: \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

FOAPAL # (19 digits) \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_\_

**Analyzer and Sorter Training Completed**: Yes or No: If this is your first-time sort request, you must complete the theory training: Email [CMtO-Core@mx.uillinois.edu](mailto:CMtO-Core@mx.uillinois.edu) for more information

**Provide a brief description of your project and your desired sort: Aria II, Melody or Bigfoot**

**\***Please note that BSL1 Yeast and bacteria are done in ARIA II and BSL2 Yeast and bacteria are done in Melody

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| ABOUT YOUR CELLS | | | | | |
| Cell Type and source.  E.g. PBMC, Mouse, Yeast, bacteria |  | | **Cell size** |  | |
| Treatment of cells (e.g., transfected, cultured, pre-enriched etc**.) If transfected, please indicate vector** |  | | | | |
| Is your samples BSL1 or BSL2? |  | | | | |
| Is this project approved for cell sorting by DRS (Division of Research Safety)? |  | | | | |
| ABOUT YOUR EXPERIMENT | | | | | |
| Staining panel and ex/em  Eg. FITC, 498/517nm |  | | | | |
| Is compensation required?  **\*If you have answered yes, please bring an unlabeled, single stained controls and a FULLY STAINED sample** |  | | | | |
| How many samples will you bring for sorting? |  | Volume of each tube | | |  |
| Number of cells present in your sample |  | | | | |
| Number of cells you would like to sort.  \*Enter a custom number or sort until the tube is empty |  | | | | |
| Have you analyzed your sample on a flow cytometer or other instrument?  What is the target sort population %?  Eg. Yes, 30% GFP+ |  | | | | |

**Tell us about your sorting experiment**

|  |  |
| --- | --- |
| **ABOUT YOUR SORTING EXPERIMENT** | |
| Temperature control during sort? |  |
| Collection tube  Eg. 1.5ml Eppendorf tubes, 5ml tubes, 96 well plate |  |
| Target populations  Eg. Unlabeled and GFP+APC-; GFP+ and APC+ |  |

**Note that all tubes, media and control samples need to be sterile for a sterile sort!**

* Cell concentration should be 1 to 5 x 106 cells per mL, however, if total cell number is low concentrate and resuspend cells in 0.5 to 1mL.
* Bring extra medium (10-20 mL) in a 15-50 mL in sterile falcon tubes for extraction/sorting and dilution if needed.
* All sorts for single cell/nuclei RNA, ATAC, Multiome and bulk sequencing requires prior analysis of samples in an analyzer (enables correct population to sort) and few test sorts before the actual sorting for sequencing. This must be done several weeks before the actual sorting of samples for sequencing.
* **Please write to** [**cmto-core@mx.uillinois.edu**](mailto:cmto-core@mx.uillinois.edu) **for an appointment or any queries.**

**Pre-sorting checklist (for your use, DO NOT fill in this section)**

Unstained control  Single stained controls for compensation

Dilution buffer (15-20mL extra)  Collection tubes with collection media

Samples have been filtered using 30- or 40-micron filters **OR** 2-3 micron for samples smaller than 2 micron

Samples have been analyzed on a flow cytometer OR microscope to confirm target population is present.

\*Please note that analyzing samples on a flow cytometer PRIOR to sorting is advised. Sorting is $196 an hour and you will be charged irrespective of the presence of your target population. Should you require training on our flow cytometers, have a look at our website.