

Tips for Sorting

1. Prepare cell samples according to your specific protocol for your cell type/application. Cells must be in single cell suspension. Live cells should be kept on ice as much as possible from the moment they are isolated until they are run on the machine. Once stained, cells should be kept in the dark.
2. Filter your prepared cells with a filter with a pore size no larger than 50µm. Be sure to filter your cells right before coming in for your appointment as certain cell types tend to aggregate and clump when left on ice for an extended period of time. Filtering right before you arrive lessens the likelihood that you will have to re-filter your material once you get here.
3. Final buffers should include 1-5% BSA or FBS to help prevent aggregation. For cells that are generally “sticky” and more likely to clump together, a small amount of EDTA can be added to help abate this problem. DNase can also be added in cases where a great deal of cell death is present or expected. Addition of DNase or EDTA should be weighed with the end goal of your sort, as the presence of either substance can be detrimental to many assays.
4. Final cell concentration should be 10-20x10⁶ cells/mL.
5. For most sorters, be sure to bring your final cell prep in 12x75 (5mL) polypropylene tubes. Cells have the tendency to stick to polystyrene and polystyrene is more prone to breakage due to the high pressures utilized by the sorters. The FACS Aria can accommodate other tube sizes such as 15mL conicals and 1.5mL eppendorfs.
6. Be sure to bring whatever tubes or plates you plan to use for collection filled with the adequate amount of media or buffer best suited for your post sort applications.
7. Each instrument has its own requirements and capabilities. Please consult each of the machine pages and the corresponding configurations in order to determine which sorter may be most suitable for your needs. Please keep in mind that human and potentially infectious samples get priority on the Aria II cell sorter and the BSL-3 samples must be restricted to Synergy cell sorter.

Feel free to contact the core with any additional questions or for consultation purposes cytometry@med.nyu.edu .