Sample Preparation for Sorting on the FACS Aria

All Runs Require:	• All the samples must be in a single cell suspension and must have been filtered through a mesh of less than 70 microns. • Samples are preferred in sterilized PBS with 1% FBS/PBS. • To avoid the clumping, DNase can be added (50ug/ml) in the final cell suspension. • Include 7AAD/PI to stain your dead cells (if your viability <75%). • For optimum sorting and save your precious samples, a diluted aliquot (300-400ul total) of your cells-to-be sorted should be placed in a separate tube in order to set up the instrument.	
Controls:	**<u>Negative unstained controls</u>** (0.3ml minimum volume)	**<u>Compensation controls for each fluor</u>** (0.3ml minimum volume)
Acquisition Samples Volume:	Cells for sorting should be in 0.5ml minimum volume if you have less than 1x10^6 cells.	Data acquisition/Record runs typically only acquire 10,000~30,000 gated events.
Sort Cell Concentration:	Samples should be at 10~20 x 10^6 cells/ml for high speed sorts and 5~10 x10^6/ml for low and medium speed sorts (5,000 to 35,000 events/s)	The Aria can sort at up to 20~100 million cells/hour (a flow rate of 0.2-0.8ul/s and up to 2ul/s with less resolution).
Collection Tubes:	Sorted cells can be accepted in 1 ml, 5 ml, or 15 ml tubes. **15ml tubes must be BD or Corning.**	The tubes should contain medium with antibiotics and 50% of FBS if healthy and viable sorted cells are preferred.
Attention:	For the first time such as particular cell sorting or preparing cells, it is best to plan a trial run of your experiment.	Be aware that if the samples are not prepared properly, then problems such as clumping and bad staining may occur; your sort may not be completed within the reserved time; and your flow profiles may not meet your expectations.
Questions:	Please direct all questions to Lisa Xu at (604) 875-2000 ext 5987 <Lixin86@interchange.ubc.ca **>**	