

BD FACSAria Quick Reference Guide

Use this quick reference guide once you are familiar with the procedures in the *BD FACSAria User's Guide*.

Starting Up the Cytometer

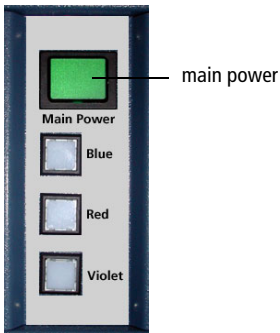
Follow these steps to start up your BD FACSAria™ cytometer. For more details, refer to Chapter 4 in the user's guide.

- 1 Start up the workstation.
- 2 Open the flow cell access door to block the lasers.



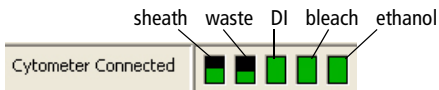
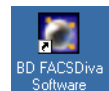
Do not allow the lasers to contact the cuvette flow cell before you activate the stream. Running the lasers without activating the stream can degrade the performance of the cuvette flow cell.

- 3 Turn on the cytometer main power and the lasers. Wait 30 minutes for the lasers to warm up.



Tip If you are using temperature control, start heating or cooling the sample injection chamber or the recirculating water bath while the lasers are warming up.



- 4 Launch BD FACSDiva™ software and log in with your user name and password.
- 5 Verify fluidics levels in the Cytometer window. Replenish fluids or empty the waste, if needed.



6 Choose Cytometer > Fluidics Startup and click OK to run startup.

7 Verify the nozzle size, then use software controls to start the stream.

As a general guideline, the nozzle opening should be at least three times the diameter of the particle to be sorted. If necessary, change the nozzle before you start the stream.

- Click the Sorting button () on the Workspace toolbar to display the Breakoff and Side Stream windows.
- Click the Stream button () in the Breakoff window to turn on the stream.

8 Check the stream position in the waste aspirator and inspect the stream image in the Breakoff window.

If you see any dripping or spraying or the stream appears abnormal, turn off the stream and refer to Troubleshooting the Stream in Chapter 7 in the user's guide.

9 Choose a new Sort Setup mode, if necessary.

Sort Setup	Nozzle (μm)	Recommended Applications
High	70	High-speed, high-throughput sorting
Medium	70	Sorting of cell lines, fragile cells
Low	100	Single-cell sorting, ACDU sorting, sorting of large or fragile cells
Custom	70	Non-sorting mode for high sensitivity

10 Adjust the Amplitude to achieve fast-merging satellites within 6 drops.

For troubleshooting assistance, see Troubleshooting the Breakoff on page 5 or refer to Sorting Troubleshooting in Chapter 7 in the user's guide.

11 Enter the generated value as the Drop 1 target.

12 Turn on the Sweet Spot when the drop pattern is stable.

Sorting Setup

Do the following to set up for sorting. For more details, refer to Chapter 5 in the user's guide.

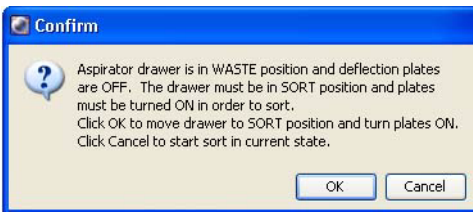
- 1 Check the laser delay and area scaling factors for your sheath pressure.
Refer to Checking Performance with Cytometer Setup and Tracking in Chapter 4 in the user's guide.
- 2 Optimize cytometer settings for the sample to be sorted.
Refer to Data Collection in Chapter 4 in the user's guide.
- 3 Install the required collection device and set up the side streams.
- 4 Use BD FACSTM Accudrop to calculate the drop delay.
- 5 Run the sample to be sorted. Use gating tools and subsetting methods to define the population(s) of interest.

NOTICE Snap-to gates cannot be used for sort gates.

- 6 Define a Sort Layout for the tube containing the defined sort populations.

Sorting

- 1 Install the collection tubes, plate, or slide.
- 2 Install the sample tube on the loading port and click Load.
- 3 (Optional) Turn on the deflection plates and open the aspirator drawer.
- 4 Verify that the current tube pointer is set to the appropriate tube in the Browser and click Sort.
- 5 Click OK if you are prompted to open the aspirator drawer or turn on the deflection plates.



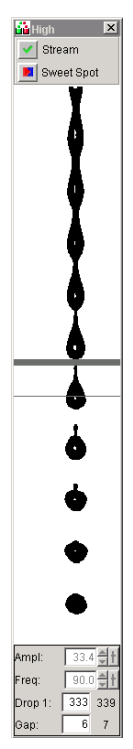
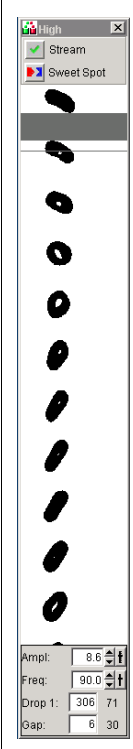
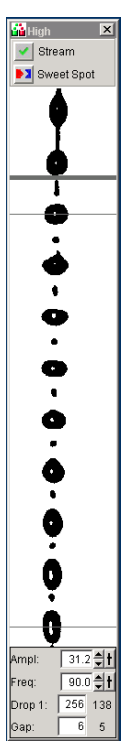
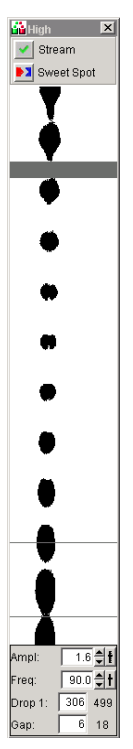

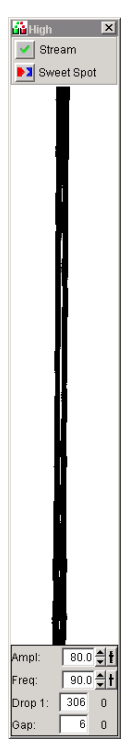
Sorting continues until the required number of cells has been sorted. If the number of Target Events is set to Continuous, sorting continues until you manually stop sorting by clicking the Stop Acquiring or Sort button.

- Tip** Click Record Data to save data for the tube. Acquisition and sorting continue when the required number of events has been recorded.

Shutting Down the Cytometer

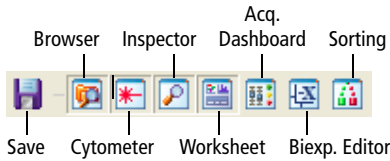
- 1** Unload your sample tube, if one is loaded.
- 2** Open the flow cell access door to block the lasers.
- 3** Turn off the stream.
- 4** Run fluidics shutdown.
- 5** Follow the prompts on the screen to shut down the fluidics system.
- 6** Turn off the cytometer main power, quit BD FACSDiva software, and shut down the computer.
- 7** (Optional) Remove the nozzle and allow it to air dry.

Troubleshooting the Breakoff

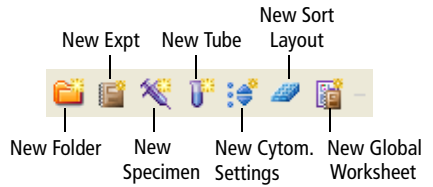
		Abnormal Stream Image					
							
Normal stream image	Possible Causes	Nozzle inserted improperly	Nozzle inserted improperly or orifice off center	Partial clog	Wet or dirty strobe lens	Attenuation on at the wrong pressure	
		Recommended Solutions	Remove the nozzle and re-insert it.	Remove the nozzle, adjust the opening, and re-insert it.	Remove the nozzle, clean it, and re-insert it.	Clean the lens as described in the user's guide.	Turn off attenuation in the Side Stream window.



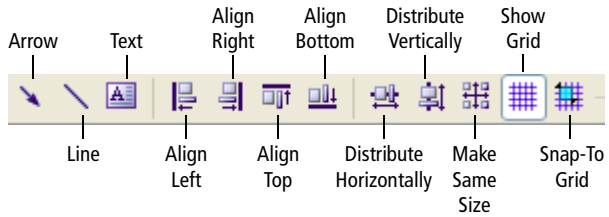
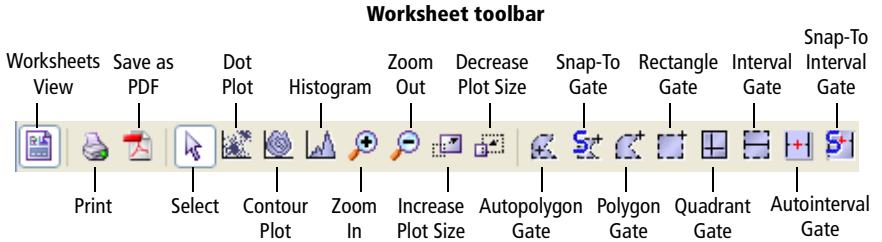
BD FACSDiva Software Toolbars



Workspace toolbar



Browser toolbar



bdbiosciences.com



BD Biosciences
 2350 Qume Drive
 San Jose, CA 95131-1807
 USA
 Tel (877) 232-8995
 Fax (800) 325-9637
facservice@bd.com

Asia Pacific
 Tel (65) 6-861-0633
 Fax (65) 6-860-1590

Europe
 Tel (32) 2 400 98 95
 Fax (32) 2 401 70 94
help.biosciences@europe.bd.com

Brazil
 Tel (55) 11-5185-9995
 Fax (55) 11-5185-9895

Japan
 Nippon Becton Dickinson Company, Ltd.
 Toll Free 0120-8555-90
 Tel 81-24-593-5405
 Fax 81-24-593-5761

Canada
 Toll Free (888) 259-0187
 Tel (905) 542-8028
 Fax (888) 229-9918
canada@bd.com

Mexico
 Toll Free 01-800-236-2543
 Tel (52) 55 5999 8296
 Fax (52) 55 5999 8288