BD LSR II User Guide

Starting up

1. **Switch on the Fluidics Cart** (located under the cytometer)
2. Turn on the power to the cytometer
3. Check the fluid levels, that the FACSFlow tank is full, and the Waste tank is empty. *In case of exchanging any of the tanks, press Restart in the Fluidics Cart right after.*
4. Switch on the computer, log in to Windows under the BDOperator user, launch and log in into the Diva Software. Wait until change to connected status.
5. Pre-run:
    a. Prime twice without tube on Sample Injection Port (SIP) (pressing PRIME in the control panel of the cytometer).

    b. Pre-run Cleaning: run FACSRinse (5min) followed by diH$_2$O (5min) while you set up for your experiment. Run at a HIGH flow rate.

    c. In case of being the first user of the day, run CS&T performance check (**Cytometer > CST**). Use an already prepared tube with CST-beads (good for up to 5 days if stored cold) or prepare a new one according to the instructions on the box.

6. The instrument is ready to run your samples.

Shutting down

1. Copy your files to an external USB (data will be deleted by the BFC every two weeks).
2. **After-run cleaning**: all washes should be done in **HIGH flow rate**:
    a. Run FACSClean with the lever open until tube is empty
    b. Run 10min FACSClean with the lever closed
    c. Run FACSRinse with the lever open until tube is empty
    d. Run 10min FACSRinse with the lever closed
    e. Run diH$_2$O with the lever open until tube is empty
    f. Run 10min diH$_2$O with the lever closed
    g. Leave a tube with diH$_2$O on the sample probe
    h. Make sure the instrument is set to Standby mode

3. Exit Diva software
4. Turn off cytometer power
5. Turn off the computer
6. Turn off the Fluidics Cart

Frequent problems LSR II

Problem: Alarm sounds on Fluidics Cart

Causes and solutions:
- **Full waste container OR Empty sheath container:**
  o Switch off the Fluidics Cart
  o Replace the full waste container by a new empty box OR get a new full BD FACS Sheath Cubitainer
  o Restart Fluidics Cart
  o Prime the fluidics after changing the tanks:
    ▪ Install an empty tube on the SIP
    ▪ Press twice the PRIME fluid control bottom
    ▪ Install a FACS tube with 1ml of deionised water on the SIP and run it on high flow rate for about 2min.

- **Waste OR sheath detectors are dirty**
  o Remove the lid of the Waste OR Sheath container and clean the detectors with a wipe and ethanol.
  o Screw up the lid again
  o Restart the Fluidics Cart

Problem: Very low or no acquisition rate

Causes and solutions:
- **Cracked FACS tube**
  o Transfer the content to a new FACS tube and rerun the sample.
  o If there is no acquisition, check for blockage or air lock.
- **System blockage**
  o Blockage needs to be shifted**:**
    ▪ Turn the instrument to Standby and try to unblock the machine by pressing the Prime buttom.
    ▪ If that did not work, run some hot water.

Problem: No events shown in the dotplots

Causes and solutions:
- **Diva Software is running slow**
  o Press Ctrl+Shift+Delete to open the Task Manager. Close the Task Manager and go back to Diva.
  o In case events are not shown, restart the computer and try again.