

SOP.UCF.038 - BD FACSYMPHONY A5 SE OPERATION

On Weekdays, if there is a slot available at 9am, the UCF staff will switch the machine ON and perform the Quality Control.

On Weekends, Holidays and outside weekly working hours (9am to 6pm), if you are the 1st user of the day you must turn ON the machine 30 minutes prior acquisition and perform the Start-up Cleaning Procedure and a Quality Control check.

1. Turn on the computer.
2. Turn on the FFSS and the cytometer.
3. Login with your Agendo Credentials:
Username – **Your email** (@*medicina.ulisboa.pt* - for internal users)
Password - **Your Agendo's password**
4. Turn on all the lasers on BD Coherent Connection 4 software (Start All).
5. Login on DIVA using your own password
6. Wait for cytometer's connection and click "**Use CST Settings**"
7. **Make sure the sheath tank is full and the waste tank isn't full before you start.** If the sheath tank becomes empty during acquisition, it will cause major problems in the equipment.

STARTING UP

- **(if you are the first user of the day and the machine is OFF)**
 8. Turn on all the lasers on BD Coherent connection 4 software.
 9. Wait 30 minutes for the lasers to warm up and stabilize while performing the daily cleaning procedure:
 10. Remove the tube with Water.
 11. Run a tube with FACSClean on HIGH for 10 min (support arm closed)
 12. Run a tube with WATER+AZIDE on HIGH for 10 min (support arm closed)
 13. Perform the Prime Check with CS&T beads.
 14. Run the Quality Control with CS&T beads.
 15. Run a tube with PBS on LOW (support arm closed) for 1 min.
 16. Run your samples.
- **(if the machine is ON)**
 17. Turn on all the lasers on BD Coherent connection 4 software.
 18. Run a tube with PBS on LOW (support arm closed) for 1 min.
 19. Run your samples.

SOP.UCF.038 - BD FACSYMPHONY A5 SE OPERATION

FINISHING ACQUISITION AND DATA HANDLING

20. **Always export your unmixed fcs files** to your Lab folder on the iMM server (shortcut on the desktop). To access the iMM server you must use your email credentials. Select the files or the experiment, right click, Export > Unmixed fcs files.
21. The first time you do the settings of a new experiment, always **export that experiment**, in case you want to redo the unmixing and/or overwrite a single color. Right-click in the Experiment to export > Export > Experiment.
22. If you have standardized settings, **export your Panel Template** to the computer. Right click in the Specimen > Export > Panel Template > On "Type" select your Lab. If you don't have standardized settings, **export your Experiment Template** to the computer. Right click in the Experiment > Export > Experiment Template > On "Type" select your Lab.
23. If you want to analyze your data in one of our Analysis Stations, you can also **export your unmixed fcs files directly to them**. To access these stations, right click on the shortcuts on the desktop (MacFlow 1 and 2) and use the following credentials:
User- flowcytometryuser Password- BDIS
24. Delete your data from DIVA as soon as you are certain that your data has been copied to the network and you have your own backup. **You cannot store experiments on DIVA for long periods of time as the software will start to malfunction. You must delete your data from DIVA and keep backups.**
25. **Clean the cytometer according to the protocol indicated on the top of the table. DO NOT FORGET TO RECORD IT**
26. **Turn off all lasers (even if someone is booked after you).**
27. **Refill the Sheath tank.**
28. If during your acquisition, **the waste container gets full**, you should put the cytometer in **Standby**, and **close** the container with the red cap that is on top of it and replace the container with a new one.

TURNING OFF THE MACHINE

After you end your experiment, please confirm in Agendo if anyone has made a reservation after you. If there you are the **last user of the day after 5pm on weekdays or the last user on weekends and holidays**, the system should be turned off.

1. Turn OFF the cytometer.
2. Sign Out of the Computer > Turn OFF the computer.