FACSymphony Troubleshooting Emory Pediatrics/Winship Flow Cytometry Core Version: 1.01 Updated: 1 March 2021

### Can't log onto Windows computer

Username: AdminPassword: BDIS#1\$\$

## Can't log into PPMS

- Check if Caps Lock is on
- Make sure there are no spaces before your username and/or password

#### **Cytometer disconnected**

- Check cytometer is powered on
- Close Diva software and reopen

## Cytometer still disconnected

- Power down cytometer (green button on side of instrument) and wait 2 minutes
- Shut down computer and wait 2 minutes
- Power on cytometer
- Power on Windows PC (see login info above)
- Turn on UV laser (see SOP for turning on UV laser)
- Open Diva and cytometer should connect automatically

#### Threshold rate is zero

- Check instrument is in **RUN** on control panel (the **RUN** button should be green if orange, the BAL seal may need replacing)
- Check for cracks in tube
- Check there is sufficient sheath and empty waste if full
- Purge sheath filter if air bubbles present
- Check all lasers are powered on in Coherent Connection software
- Check for clog (run a tube of water on **HI** and make sure fluid level goes down)
  - o If fluid level does not change, run the **PRIME** function on control panel
  - o Run water tube for 5 minutes to remove any air bubbles from flow cell
- Check that threshold is set to the correct parameter (typically FSC)
- Check that threshold is not set too high (typically 5,000)
- Check PMT voltage for threshold parameter is not set too low
- Try making a new experiment

# Tube does not stay on SIP without support arm in place

- Check for cracks in tube
- Check that proper tubes are being used:
  Falcon polystyrene (clear) tubes: Fisher cat # 352008

#### UV dyes not detected

- Check UV laser is powered on and set to 60 mW
  - Open Coherent Connection/UV laser software (should be minimized in Windows taskbar)
  - o If Coherent Connection software is closed, double click on the icon to open it

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- Note: Opening the software will automatically turn off the UV laser
- Turn on UV laser (see SOP)
- Minimize Coherent Connection software DO NOT CLOSE