Research Flow Cytometry Facility

Correct

Not Correct

Troubleshooting Guide

Problem: No/Few events being seen during acquisition

1) Is the green run light on?

a. Yes

1. Is your tube selected?
   - Arrow next to tube name should be highlighted green
2. Try a new experiment
   - Templates can unexpectedly become corrupt
3. Is the instrument connected properly?
   - Sometimes a "9" event threshold occurs
     a. Turn off computer and instrument then turn on computer first and then instrument (and UV laser for Fortessa 1)
4. There might be bubble in flow cell
   - Prime 2x with nothing on sit (air bubbles should be coming out)
5. If none of the above fixes the issue, proceed to the step below.

b. No

1. Check/Refill tanks
   - Make sure o-ring is in place and all lines are connected correctly
   - Debubble sheath tank filters and side filter
   - Waste line may not be plugged in properly
2. Is your tube cracked?
   - Get new tube
   - Is your tube a 5 ml BD falcon **polystyrene**?
     a. Change your tube to Catalog # 352058, 3502052, 352008, 352235
3. Are samples filtered and free of floaties?
   - Filter ALL samples prior to running
   - If running for longer than 1 hour, it is recommended to run a 3 minute cleaning of bleach and water.
4. Are your cells at the correct dilution?
   - 1-5 x 10^6 cells/mL is suggested
     a. If not at correct dilution, dilute out sample with buffer or sheath
5. Instrument may be clogged.
   - Run 100% hot bleach for 3 minutes on HIGH followed by 5 minutes of H2O on HIGH

If this fails, please call 6-3573 or visit the lab, R5503. You can also text Alyssa @ 859-743-3323 or find any one of the RFCF Lab Members.