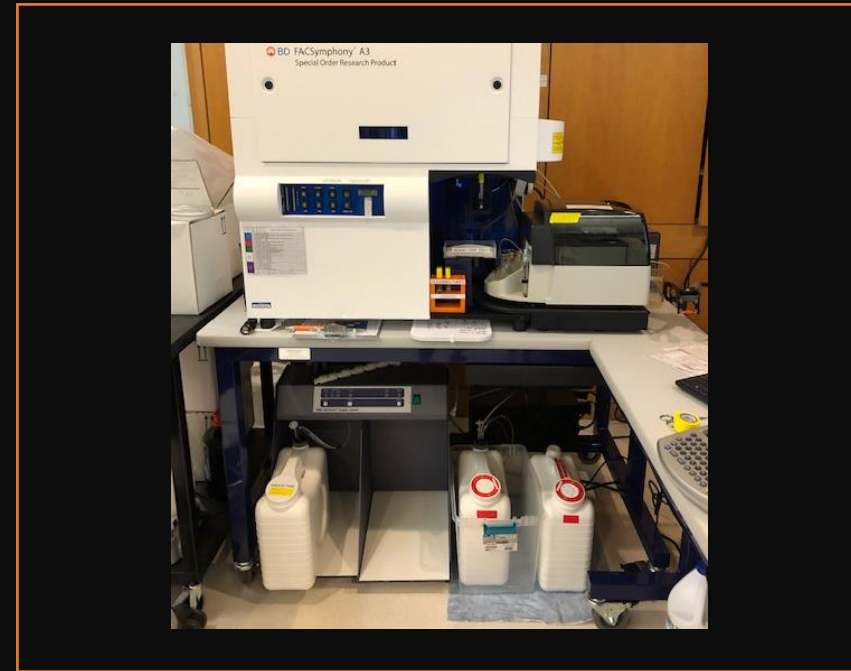
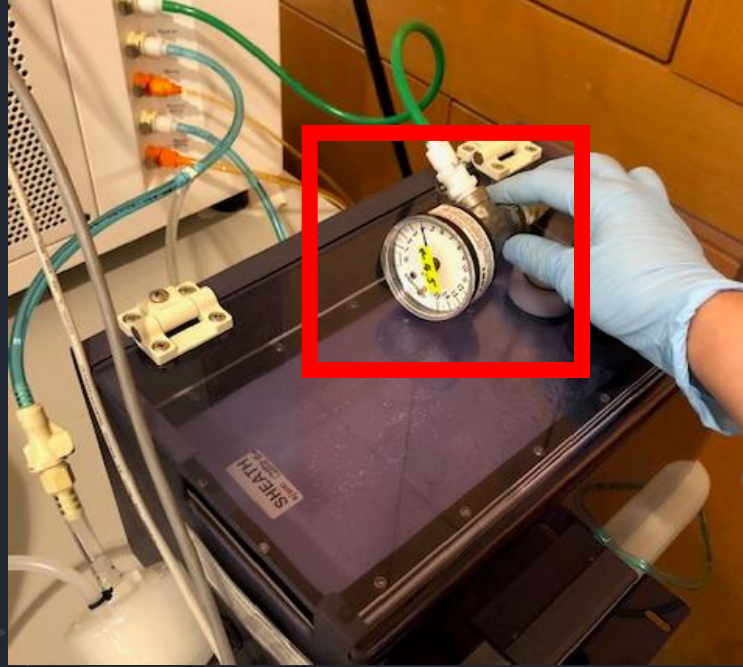
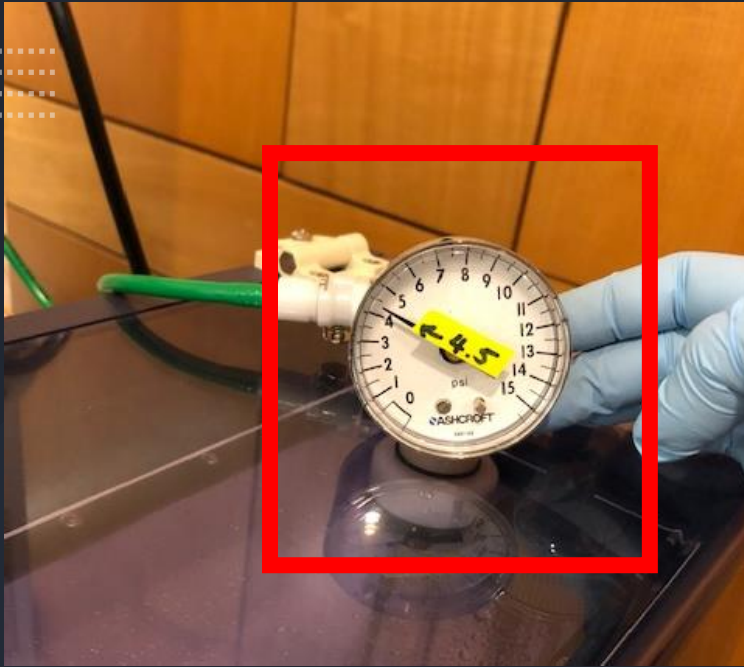

Troubleshooting Guide for A3 - Run Button Is Yellow

- When the run button is yellow, this is indicative of a fluidics pressure issue somewhere in the A3 system. To troubleshoot the issue, start at the beginning of the A3 instrument fluidics (the fluidics cart sheath tank, i.e. sheath tank filled) and move through to the end (the waste tank, i.e. waste tank emptied). The following slides will help you through this process in a step by step fashion.

1. Check that Fluidics Cart is ON

- Fluidics Cart is located directly below A3. When ON, the “System On” light and the green Fluidics Cart power button will be lit up (as indicated by red box in picture above)
- If that is not the case, then switch the Green Power Button to “ON” position.



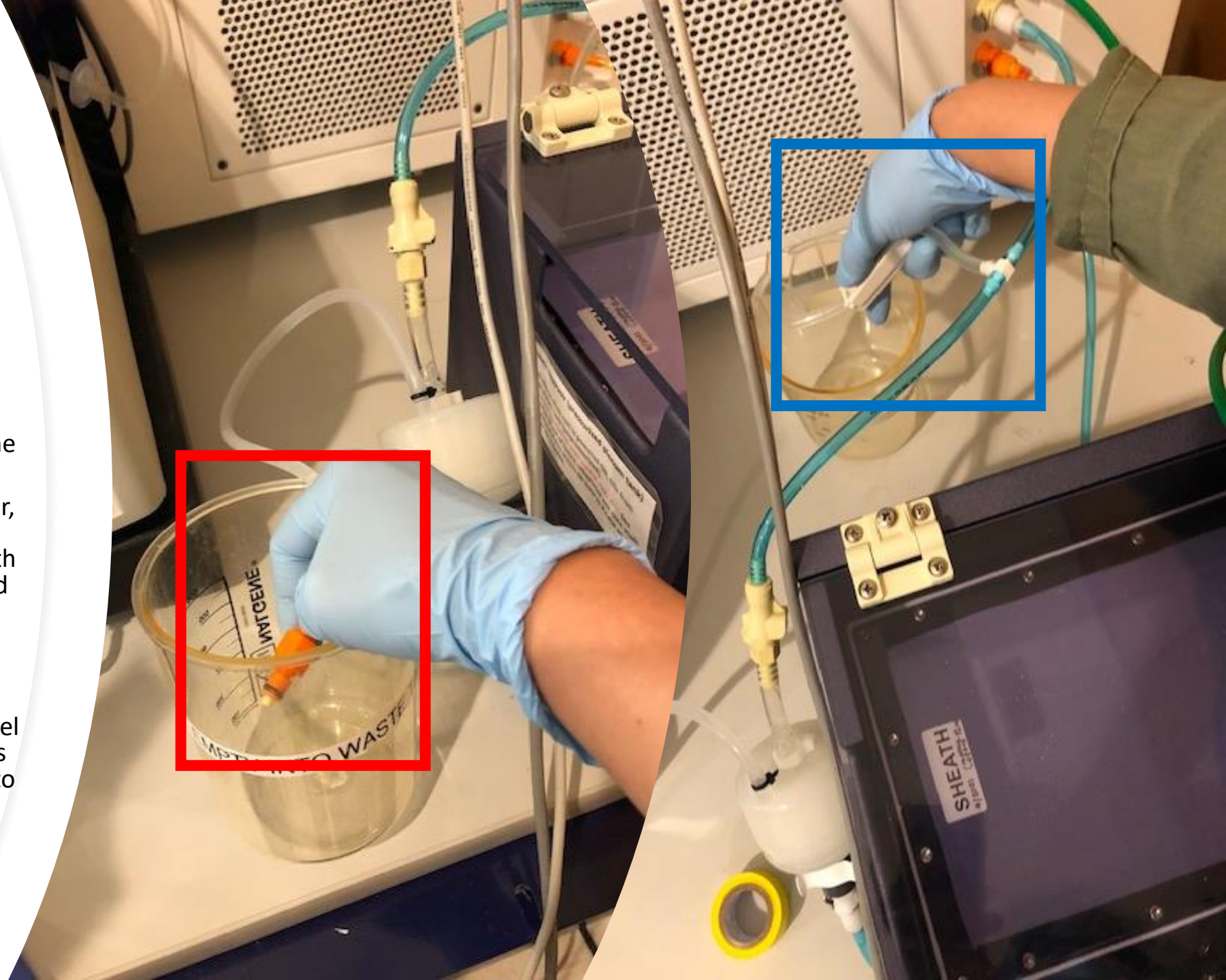


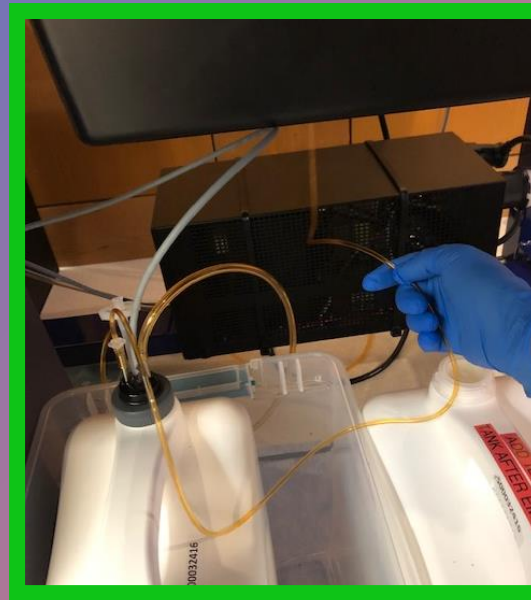
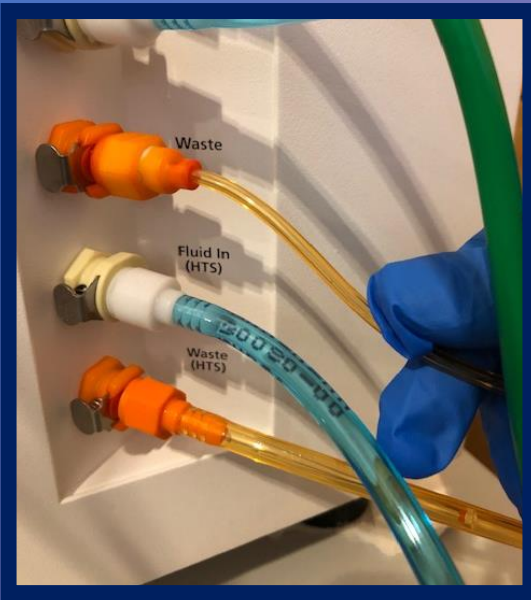
2. Check that plenum is pressurized and has sheath
plenum tank is encased in the gray box located to the right of the A3 on A3 instrument table

- There is an air pressure gage that we keep placed on top of plenum tank box. Check that this pressure gage (indicated in the photos above by the red boxes) reads 4.5 PSI.
- Check that the fluid level within the plenum is at least 1 inch high by using flashlight. **The plenum itself should never be empty, and should be maintained less than 2 inches by pump).** At this point, you should also check that the fluid levels in the sheath and waste tanks are appropriate to continue.

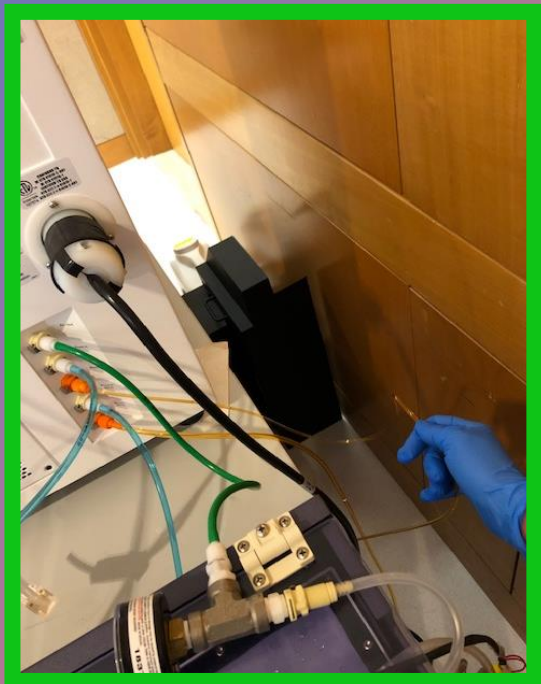
3. Check sheath filter for air/bleed sheath filter

- If the instrument is run dry, air can get into the sheath filter and into the sheath/waste lines. If that is the case, then the air must be removed to continue.
- First, check the sheath filter located to the side of the plenum box.
- If there are air bubbles in the sheath filter, bleed the sheath filter by using roll valve (indicated by blue box) and bleed tubing with orange quick release fitting (indicated by red box). Bleed any air into a plastic container located on parts cart or next to A3 (empty fluid into waste tank afterwards).
- To use roll valve, roll wheel away to open releasing air/fluid, then close by rolling wheel back to original position. Make sure wheel is closed tightly, so sheath does not continue to leak out.
- Repeat these steps as necessary: Trace along the tubing and ensure there are no visible dry sections in the tubing as it runs **from the plenum > to the sheath filter > to the A3 side panel**





- Once the instrument run button turns green (and it appeared that the instrument may have been run dry), start running a tube of water and while the water is running, inspect the orange waste tubing for stationary bubbles that may have accumulated. Fluid in the waste tubing should move smoothly to the waste tank: if stationary bubbles are visible, give the orange tubing a couple of gentle flicks to dislodge. **Take care not to handle the tubing near any connections to the instrument or waste tank as these could snap off.** Examples of where to hold waste tubing shown in pictures with green perimeter.



- You can identify the waste line by looking at the labelling where the quick disconnect fittings connect into the A3 side panel (indicated in picture with blue perimeter)
- An example of the air bubbles in waste line are shown in the picture indicated by the pink perimeter.

4. Check that your FACS tube is not cracked.

- If the run button is still yellow, and none of steps 1-3 have resolved the fluidics pressure issue, check that your tube is not cracked. If your tube is cracked, it cannot be fully pressurized to take up sample (also your tube may quickly fill up with sheath when installed if this is the case).
- Remove the tube from the SIP, and check the integrity.



5. Contact FCCF, use another analyzer if possible

- If you have followed all the steps in the previous slides, but the RUN button is still Yellow, contact FCCF@salk.edu or find an FCCF staff member to help you further troubleshoot the issue.