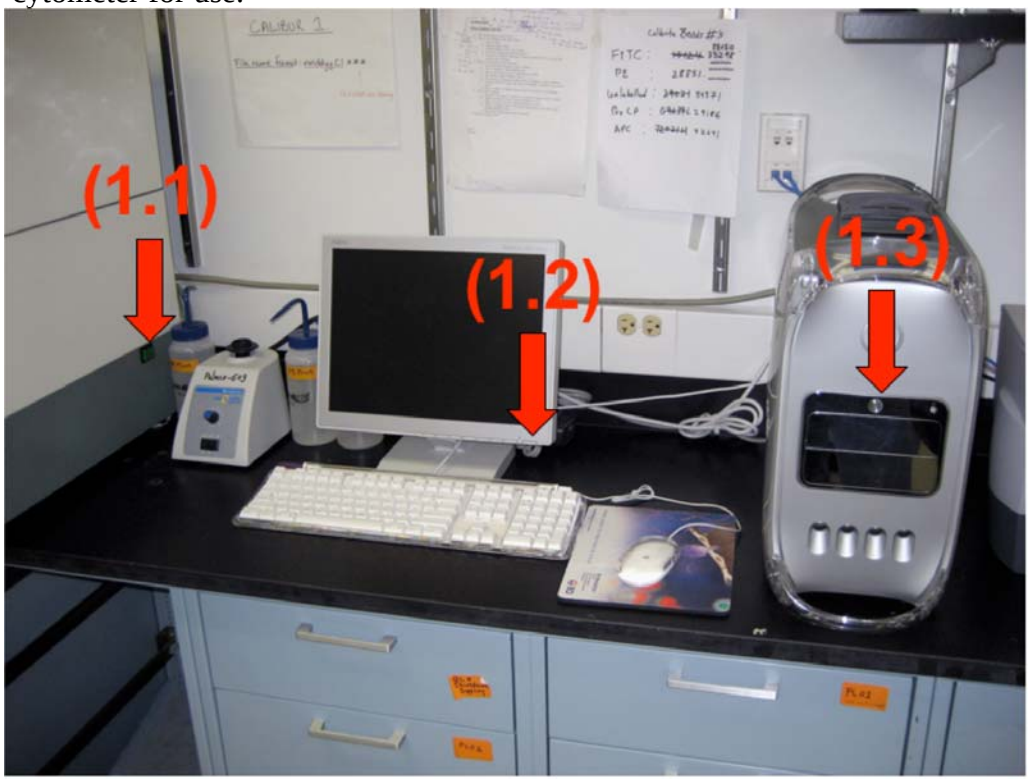
BD FACS Calibur Instructions

The BD FACS Calibur is available to all investigators who have received ImageCORE flow Cytometry training and desire the quantification benefits of immunocytometry. The system is QC'd on a weekly basis in order to track instrument reliability and help highlight any potential problems in the system. More frequent QC's are available to any investigator at an additional cost per test.

Power Up Procedure

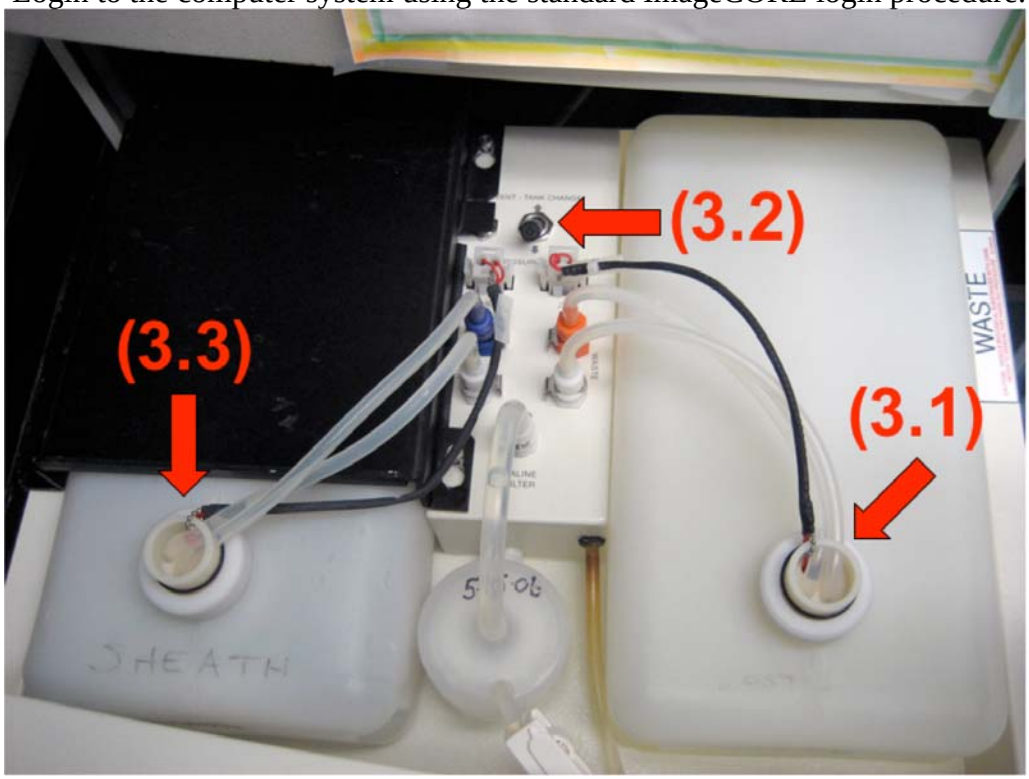
- All peripheral devices must be powered on prior to starting the flow cytometer computer workstation.
- Power on the flow cytometer (1.1), the computer monitor (1.2) and the computer system (1.3). It is also necessary to check that the printer is on (should be left on at all times).
- Once all systems are powered on and running, it is necessary to prep the cytometer for use.



- Confirm that the cytometer control buttons (2.1) are set to “LO” and “Standby” prior to opening the fluidics drawer.
- Open the fluidics drawer (2.2) and obtain a few paper towels to assist in the instrument prep.

- Begin prepping the instrument by emptying the waste container (3.1). This will be the only container removed from the system during the prep. Remove the cap from the waste tank being careful to not pull on the electronics attached. Place the cap on top of a paper towel (obtained earlier) out of the way of the waste tank.
- Remove the waste tank from the fluidics drawer and drain into sink with hot water running.
- Wipe the waste container dry after emptied and return to the fluidics drawer.
- Locate industrial bleach under the sink and fill the waste container with 200ml of bleach.
- Refit the cap to the waste container making sure to snug the cap tightly and orient the electronics towards their respective connections in the center of the fluidics drawer.
- Purge the pressure from the sheath fluid tank by switching off the vacuum with the purge valve (3.2).
- Remove the cap from the sheath fluid tank (3.3) and again be careful of the electrical connections. NOTHING will need to be unplugged. Place the cap aside on top of a paper towel (obtained earlier).
- Locate the sheath fluid reservoir on the shelf directly above the sink. It is not necessary to remove the sheath fluid tank. Refill the tank to the top indentation (3/4full) using the hose attached to the sheath fluid reservoir.

- Once filled to the top indentation, refit the sheath fluid cap and snug, taking care to orient the electronics towards their respective connections.
- Reengage the vacuum system via the purge valve (3.2). Confirm that pressure in the system is sustained by briefly changing the cytometer control buttons (2.1) to "Lo" and "Run." The "Run" light should change from an initial color of amber to green. If a green "Run" light is not obtained, check that both caps on the waste and sheath fluid tanks are snug as well as that all the fittings in the center of the fluidics drawer are connected.
- Return the cytometer control buttons to "Lo" and "Standby."
- The cytometer is now prepped and ready for use.
- Login to the computer system using the standard ImageCORE login procedure.



Shutdown Procedure

- Obtain three 3ml FACS tubes. Fill two with 2ml DDI H_2O and another with 2ml 10% Bleach. All DDI and Bleach is provided in labeled blue squeeze bottles located next to the flow cytometer.
- Set the cytometer control buttons (4.1) to "High" and "Run." Place the tube with 10% bleach on the sipper of the cytometer. Make sure to leave the arm off to the side in the non-engaged position (4.2). You will hear a noticeable vacuum sound and the sample will visibly be removed from the tube.
- After 1 minute has elapsed, engage the sample arm into position (4.3). Leave the sample on for an additionl 5 minutes as the bleach flushes the flow cell and subsequent flow lines.
- Repeat the same procedure (1 minute arm disengaged, 5 minutes arm engaged) with one of the DDI H_2O tubes.

- Having completed the flushing of the cytometer, it is necessary to place the second tube of DDI H_2O on the sipper and engage the arm (4.3).
- Set the cytometer control buttons (4.1) to “Lo” and “Standby.” You may now power off the cytometer and shutdown the computer system.