Aurora Operation

URMC Flow Cytometry Shared Resource Lab

March 2022

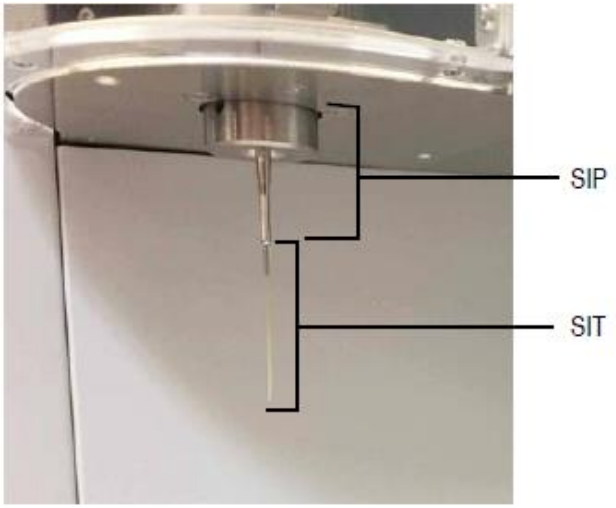
The purpose of this document is to familiarize the user with the fluidics and operational components of the Cytek Aurora. This is meant to be a basic operational guide and does not cover troubleshooting or the Spectroflo software.

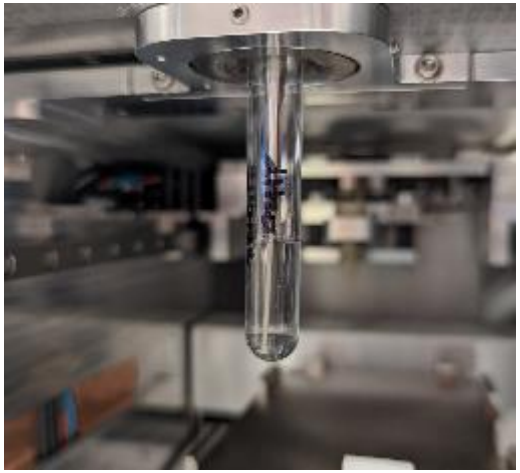
The SIT and the SIP

1. The SIP is where tubes are attached to run samples
    - The Aurora's SIP fits standard 12x75mm 5mL tubes, the same as the BD instruments.
    - Tubes on the Aurora do not need to create an airtight seal in order to run sample; the Aurora uses vacuum pressure to move sample and sheath fluid through the fluidics pathway.
    - Tubes snap into place on the SIP and are held with mechanical force. Only gentle pressure is needed to place the tube onto the SIP. This will be accompanied by an audible click when the tube is attached.

2. The SIT is the piece of tubing that descends into the sample tube and delivers the sample to the flow cell for interrogation at the laser intercept.
    - When there is no tube on the SIP and no sample is being run, the SIT is retracted into the SIP.
    - When a user places a tube on the SIP and begins to acquire sample the SIT will descend to just above the bottom of the tube and begin to draw the contents of the tube into the machine.
    - It is important to remove tubes from the SIP **AFTER** the SIT has retracted. **Removing a tube before the SIT has fully retracted may result in the acquisition tubing becoming bent.**

3. Every time a tube is taken off of the SIP the Aurora performs a SIT flush function. This is accompanied by a small window that pops up in the software that displays a progress bar for the SIT flush. Additional flushes can be run as desired by clicking the button in the Acquisition Control window.

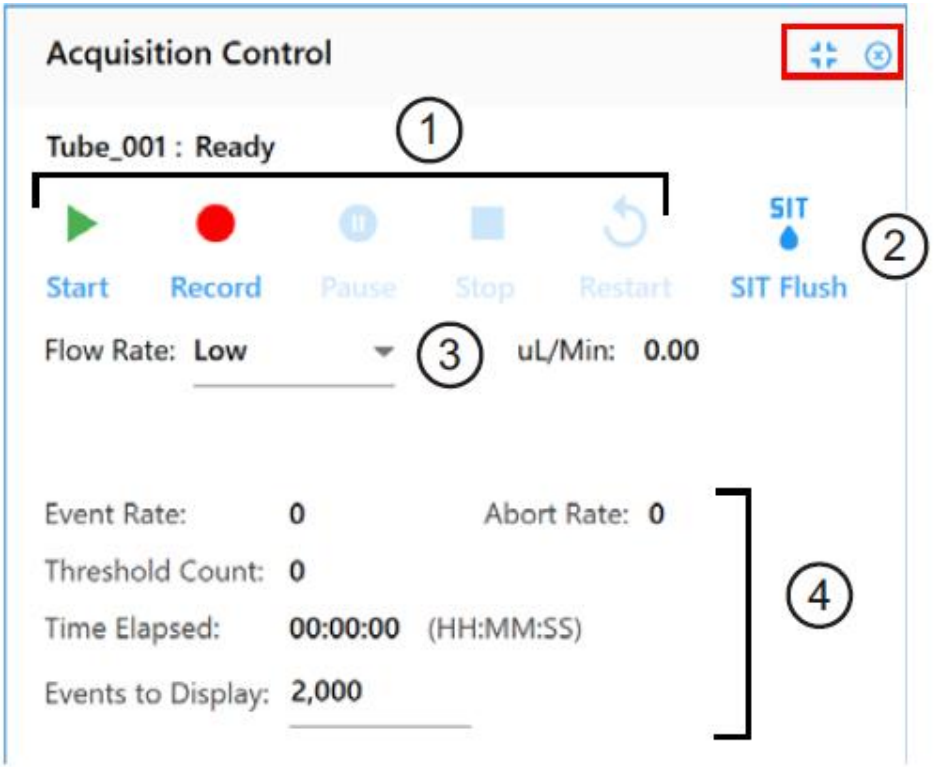




Example of sample being acquired from a loaded tube

Acquisition Controls

The acquisition controls are located in the 'Acquisition' portion of the Spectroflo Software



1. Start/Record/Pause/Stop/Restart
    - Start: Enabled when a tube is present on the SIP. Starts sample acquisition
    - Record: Enabled when a tube is present on the SIP. Records data. The record button can also begin acquisition
    - Pause: Pauses data recording. Pausing will allow the user to adjust the flow rate during acquisition. Select Record after pausing to continue recording
    - Stop: Stops acquisition.
    - Restart: Restarts the acquisition counters (4) and refreshes all events and results displayed
2. SIT Flush
    - Performs a SIT flush if additional SIT flush is desired between or after samples
3. Flow Rate
    - Opens a dropdown menu to select flow rate. Users can select Low (~15uL per minute,) Medium (~30uL per minute,) or High (~60uL per minute.) The exact flow rate is displayed as samples are being run.
4. Event Rate/Abort Rate/Threshold Count/Elapsed Time/Events to Display
    - Event Rate: Displays the rate at which events are being run through the instrument
    - Abort Rate: Displays a count of aborted events
    - Threshold Count: Displays the number of events counted by the instrument since beginning or restarting acquisition/recording
    - Elapsed Time: Displays time elapsed since beginning or restarting acquisition/recording
    - Events to Display: Allows user to set the maximum number of events that are displayed on plots in the worksheet.

The Sheath Supply

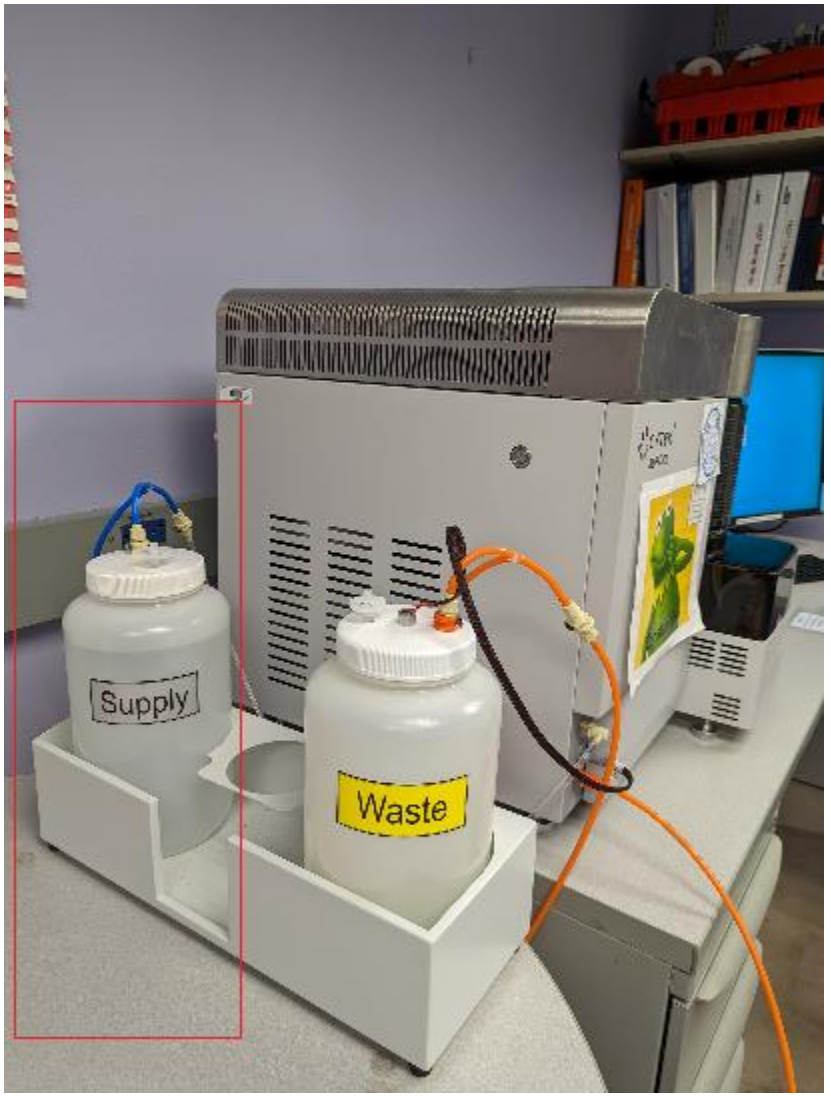
The Sheath Supply bottle is located in the small white stand to the left of the instrument and it is labeled 'Supply' for clarity.

Sheath fluid for the Aurora is ultra pure filtered water with no additional additives.

The Spectroflo software will warn the user when the sheath supply is low or empty by displaying a yellow (low) or red (empty) fluid indicator icon in the lower right corner of the software window.

To refill the supply bottle, first make sure the instrument is not running any sample or cleaning fluid. Disconnect the quick-connect fitting on the lid and take the bottle to the Gemini water system. Remove the lid and dispense ultra pure water into the supply bottle until the liquid level reaches the shoulder of the bottle. Replace the lid of the supply bottle and reconnect it to the instrument.

The supply bottle does not pressurize and is not subjected to vacuum pressure while the instrument is running.



The Waste Bottle

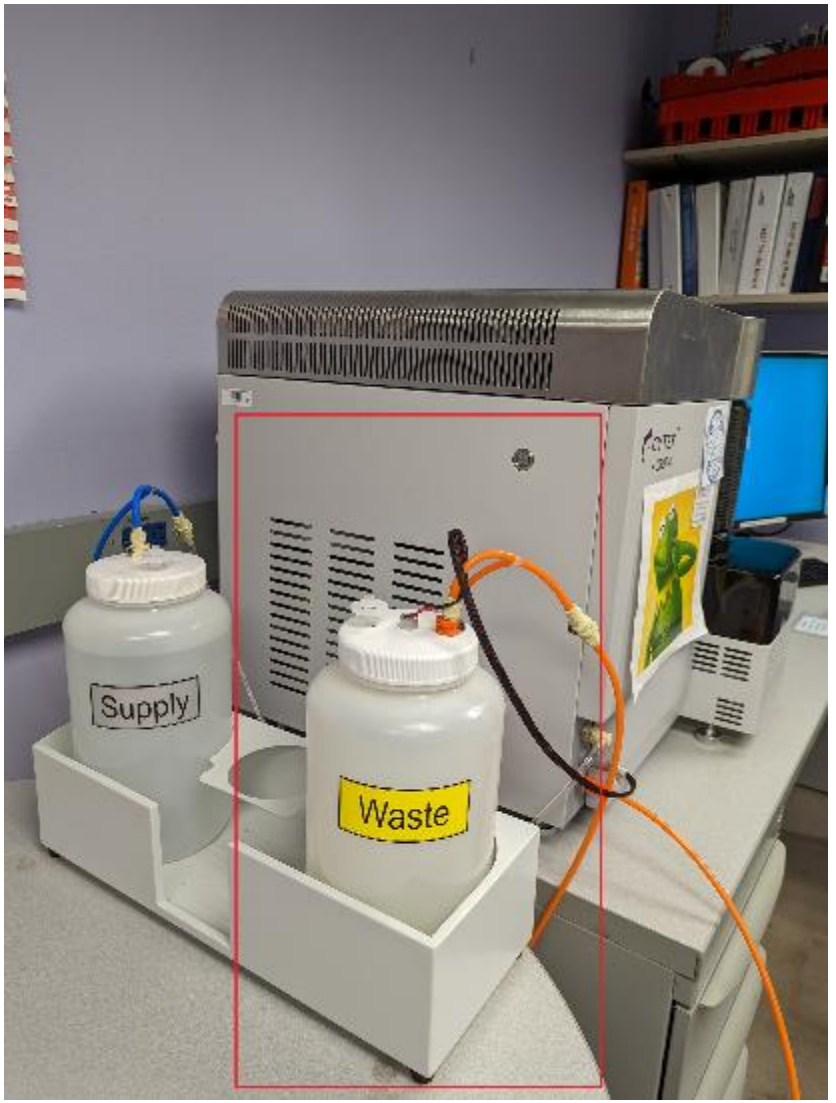
The Waste bottle is located in the small white stand to the left of the instrument and is labeled 'Waste'.

The software will prevent further acquisition and prompt the user to empty the waste when the waste sensor detects that the bottle is full. It is not necessary to wait for the prompt in order to empty the waste bottle.

The Spectroflo software will warn the user when the waste is nearing capacity or full by displaying a yellow (nearing capacity) or red (full) fluid indicator icon in the lower right corner of the software window.

To empty the waste, disconnect the quick-connect fitting on the lid of the waste bottle and pour the contents down the drain of the sink in the adjoining laboratory. Run water from the tap for approximately 15 seconds after pouring the waste out. Before replacing the lid, pour bleach (located beneath the sink) into the bottle until the level of bleach is approximately 2cm above the bottom of the bottle. Reconnect the waste bottle to the instrument.

The waste bottle is not pressurized and is not subjected to vacuum pressure while the instrument is running.



Important Things to Remember

- **Before you begin**: Check the waste and sheath supply bottles to be sure your run is not interrupted. Always fill the sheath supply bottle if you empty the waste to make sure they are in balance.

1. **Normal Cleaning**: Perform a 'Clean Flow Cell' function. Can be accessed in the 'Cytometer' menu located in the leftmost column of the Acquisition and QC modules. A prompt will appear that will guide you through the process.

2. **Shutdown Cleaning:** Perform a 'Fluidics Shutdown' function. A prompt will appear to guide you through the process. At completion leave the water tube in position and power off the instrument.