FSU | COLLEGE OF MEDICINE

STANDARD OPERATING PROCEDURE

Operation of Aurora

1. Purpose and Policy

The purpose of this document is to provide standard for best practices when using the Aurora analyzer for flow cytometry experiment.

Aurora offers full spectrum technology and auto fluorescence extraction in SpectroFlo software. Spectral unmixing feature allows the use of two dyes with similar peak emissions. It equips with 5 lasers (355nm, 405nm, 488nm, 561nm, 640nm) and total 64 APD detectors enable more efficient spectrum capture for dyes emitting in the 365-829 nm range.

The following procedures are listed in the Cytek Aurora User's Guide. The SpectroFlo software also provides complete guidance during all the procedures. This SOP should not be considered to replace the User's Guide or guidance from software.

The Aurora can be operated by users if proper training is received and proved by lab staff.

2. Procedure for User

Users are encouraged to discuss with lab staff about panel design, protocol optimization and instrument settings before experiments.

If any issue is found by the user during the experiment, it should be reported to the lab staff immediately.

Preform the step 2.1 and 2.2 if you are the first user of the day. Preform 2.4 if you are the last user of the day.

2.1 Instrument Startup

- Check the level of sheath fluid (ddH_2O) and waste. Fill the sheath tank and empty the waste tank as needed.
- Turn on the Aurora and the ALS loader. Log in the computer with user's MED ID.
- Start the SpectroFlo software and log in with user's ID.

- Wait for the loader to move to the eject position, remove the tube rack.
- Load a tube of 3 mL ddH_2O in the SIP and wait for the instrument to start up.
- Confirm the status lights (sheath, waste, cytometer, loader) are all green.
- Open a default experiment, run ddH_2O as sample for 30 mins.

2.2 QC

- Confirm the lot of SpectroFlo QC beads. Vortex the tube well.
- Add one drop of the beads into 300 µL of ddH_2O and vortex. Label the tube and date.
- From the QC tab of the software, verify and select the QC beads lot.
- Load the QC tube and start acquisition. Wait for the QC procedure to be completed.
- Store the new diluted tube in fridge. Discard the diluted QC tube after a month.
- Pass/Fail Criteria:
  %rCV must not exceed 6% for the FSC channel
  %rCV must not exceed 8% for the SSC and SSC-B channels
  %rCV must not exceed 6% for the third channel of each laser (V3, B3, R3, YG3, and UV3)
  % delta gain change for all channels must not exceed 100% from the last Daily QC run performed by Cytek Service personnel.

2.3 Instrument Shutdown

- Prepare four tubes of cleaning solutions (10% bleach, ddH_2O, 30% Contrad, ddH_2O, at least 3 mL each), and place in the A1-D1 position on the tube rack.
- Start the "**Fluidics Shutdown"** and wait for the procedure to complete.
- Turn off the Aurora and the ALS loader. Close the software and log out the computer.

2.4 Data Management

- It's user's responsibility to keep all the flow data (FSC file, batch analysis, experiment settings). The data on the computer may be deleted without notification.
- Experiment data should be only transferred through user's OneDrive or Med folder. Using thumb drive is prohibited. Ask lab staff for help if you don't have an account.

3. Special Procedure for BSL-2 Requirements

Follow this procedure if your sample requires BSL-2. Read and sign the BSL-2 procedures. Notify the lab staff before run the experiment.

3.1 Sample Preparation and Liquid Handling

- Sample preparation should be done in a BSL-2 hood in the user's lab and transfer to flow lab in a close cap tube.
- The tube cap should keep close all the time when vortex the tube.
- Centrifugation can be performed only with the aerosol-tight caps.

3.2 Decontamination

- Before starting the experiment, empty the waste tank and fill with 200 mL 100% bleach.
- After running all the samples, run 3 mL 10% bleach sample for 10 mins, and proceed to Fluidics Shutdown as in step 2.3.
- Wipe all the surface, loader and bench with 70% alcohol.

3.3 Waste

- Solid waste (tips, tubes) should be submerged in 10% bleach and autoclaved following BSL-2 procedure.
- Leave the waste tank with 10% bleach (final concentration) for at least 30 mins.

4. Procedure for Lab Staff

4.1 Maintenance

- Lab staff should confirm the instrument is shut down at 5 pm on the workday.
- **"Aseptic Clean/Long Clean"** procedure should be performed every month, and anytime necessary.
- The filter on the sheath and waste tank should be replaced every 6 months, or as needed.
- Sheath filter should be replaced every year or as needed.
- Instrument QC should be done at least once a month if not frequently used.
- All the maintenance procedures should be recorded, including septic cleaning, filter change and data backup.

4.2 Data management

- The experiment data on the computer should be backup to the FACS Data drive using the backup tool in the software once a month.
- All experiment data will be deleted if free space on the C drive is less than 30%.

5. References

Cytek Aurora User's Guide (N9-20006) is stored in the folder on the desktop for reference.

SpectroFlo software.

Also see the ISAC SRL (https://archive.org/details/@isac_srl) for more protocol and SOPs.

BSL-2 Facility Policies and Procedures.

6. Approvals

The signatures and dates below are required for full SOP approval and implementation.

This SOP was written by:

Name	Title	Signature	Date
Li Sun	Director	Li Sun	7/24/2026

This SOP was reviewed and authorized by:

Name	Title	Signature	Date
Li Sun	Director	Li Sun	7/24/2026