

General

Registration with the Flow Cytometry Facility, as well as attending the technical introduction by the facility staff is a prerequisite to use the Cytek Aurora.

Questions / reporting technical problems:



Feedback form

Emergencies:

☎ 044 635 02 03

☎ 044 635 02 06

The loader works with:

- Standard 96 well (U-, V- or flat-bottom) plates.
- Nunc and Brand 96 deep well plates (1, 1.3, 2 and 2.2 ml)
- FACS tube rack (40 positions)

Differences to the loader on the Aurora 1:

- Loads tube rack and deep well plates.
- No cooling or heating.
- Cell resuspension via shaking of the stage.



	96 U-Bottom
	96 V-Bottom
	96 Flat-Bottom
	Nunc 1.3mL 96 Deep
	Nunc 2mL 96 Deep
	Brand 1mL 96 Deep
	Brand 2.2mL 96 Deep
	40 Tube Rack

I. Setting up the loader

1. **Make sure that there is no tube on the SIP and that the sample line is pulled into the instrument.**
2. **When using plates:**
→ Pull the lever to the front.
When using the tube rack:
→ Leave the lever in the back position.
3. Switch on the plate loader via the switch at the back.
4. Wait for the Loader status display in the software to turn green:

Loader



5. Go to Preferences → Cytometer and make sure that the “Enable Bubble/Clog Detection” is turned on in the Fluidics section. Choose what should be done in case of bubble or clog detection.
6. If plate tray is not at the front, click the Eject button in the Cytometer tab.
7. Put your plate or rack on the tray. **A1 position is at the front left.**
8. Load the plate/rack (move it to the back of the loader) by clicking Load in the Cytometer tab.
9. Go to the Cytometer tab and perform a Calibrate SIT. Select the appropriate plate or rack and set the Lift Distance (see below).
10. The loader is now ready to use.

Fluidics

Skip Fluidics Boost when Starting Acquisition for High and Medium Flow Rates

Enable Bubble/Clog Detection

In Case of Bubble

Move to next target after SIT flush

Stop immediately after SIT flush

In Case of Clog

Move to next target after SIT flush

Stop immediately after SIT flush

II. Verify SIT calibration

This step is recommended whenever you use a certain type of plate for the first time.

Note: Lift distance settings from other Auroras are not necessarily transferrable between instruments.

1. Fill a well with ddH₂O and load the plate.
2. Acquire from the well and check the flow rate.
Expected flow rates in plate mode are:
 - Low: 15 ul / min
 - Medium: 30 ul / min
 - High: 100 ul / min
3. If the measured flow rate is too low or you get a “Clog detected” message, go to the Cytometer tab and perform a Calibrate SIT with a higher Lift Distance. Standard value is 0.75.
4. Repeat steps 2 and 3 until you get the desired sample flow rate.

III. Set up plate/rack in your experiment

1. Create a new experiment or edit an existing one.
2. Change carrier type to tube rack or the plate type you are using.
3. Under Groups, add the plates you want to acquire.
4. Select the appropriate positions and click the buttons on the right to create groups for normal samples, reference controls or cleaning wells/tubes.

Plate_001 96 U-Bottom

	1	2	3	4	5	6	7	8	9	10	11	12		
-													+	Sample Group
A	○	○	○	○	○	○	○	○	○	○	○	○	R	Reference Group
B	○	○	○	○	○	○	○	○	○	○	○	○		
C	○	○	○	○	○	○	○	○	○	○	○	○	C	Cleaning Wells

5. Set the loader settings. Choose from Default, High Throughput or Low Carryover or create your custom settings.

Loader Settings

Plate_001

Low Carryover (System) Save

⇌
⇌

Shake Time	Shake Speed
2	1300
Shake Interval Mode	Shake Every N Wells
Well	8
Premix Time	SIT Flush Times
3	Double Flush
Sample Recovery	Record Data Delay Time
Off	0

Acquisition order	<ul style="list-style-type: none"> row from left to right (A1-A12, B1-B12, etc) column from top to bottom (1A-1H, 2A-2H, etc) row from left to right, then right to left (A1-A12, B12-B1, C1-C12, etc) column from top to bottom, then bottom to top (1A-1H, 2H-2A, etc)
Shake Time	Select the time (in seconds) to shake the plate/tube rack. You can also disable shake time.
Shake Speed	Select the speed of the orbital shaker (in RPM).
Shake Interval Mode	Select whether you would like to shake every N wells or after a specified period of time. You can also disable shaking.
Shake Every N Wells, or Shake Interval	Select how often (number of wells or time in seconds) to shake the plate/tube rack.
Premix Time	Select the time (in seconds) to shake the plate/tube rack before acquisition of the first tube/well.
SIT Flush Times	Number of SIT flushes performed after each acquisition.
Sample Recovery	Allows any remaining sample that is left in the SIT after acquisition to be deposited back into the well.
Record Data Delay Time	Time in seconds before recording starts (to e.g. exclude the boost phase).

IV. Change back to manual tube loading

Please always change back to manual loading mode at the end of your session to prevent problems for users not familiar with the loader.

1. **Make sure that the sample line is retracted into the instrument.**
2. Remove your plate/rack from the loading tray.
3. Click on "Load" in the Acquisition Control or in the Cytometer tab to move the tray to the back.
4. Switch off the plate loader.
5. **If plates were used:** Move the lever to the back.
6. Change the carrier type back to manual tube loading.
7. Run the cleaning procedure as described in the Cytek Aurora SOP.