Cytoflex plate reader guide (Sep 2026)

Machine power on

Perform setup and QC in tube mode (semi-automatic)

Check sheath and waste tanks

Ensure tanks won't need attention during the plate run

Put into plate mode

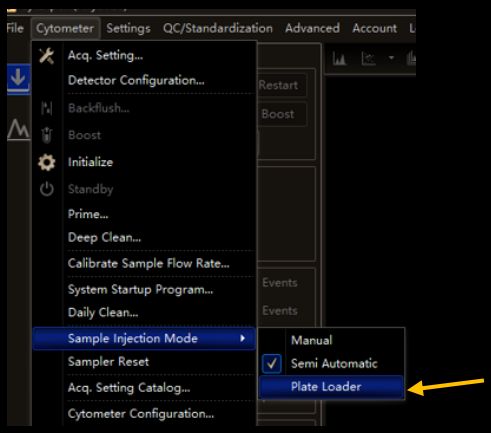
Check if in semi-automatic (tube) or plate mode

See next page for tube > plate instructions

Cytoflex: Changing sample injection mode

Tube → Plate mode

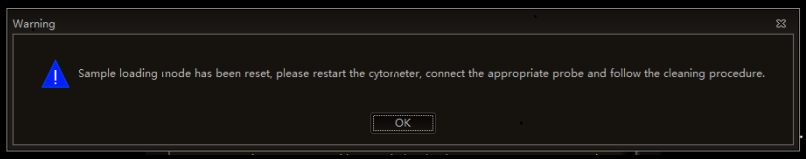
1/ [Cytometer] > [sample injection mode]



2/ Select [plate loader]

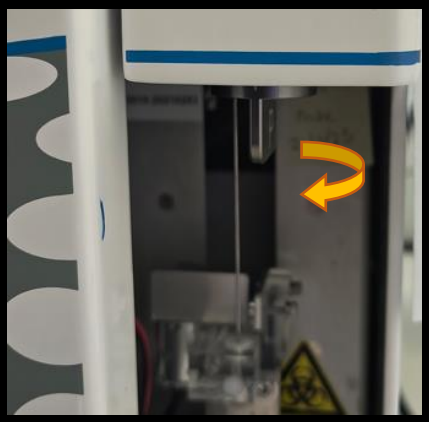
3/ Dialog box will appear

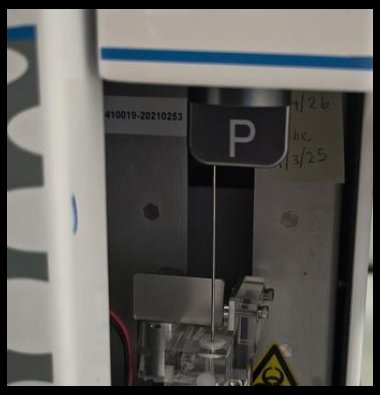
Sample loading mode has been reset, please restart the cytometer, connect the appropriate probe and follow cleaning procedure



4/ Power off the Flex at the power point

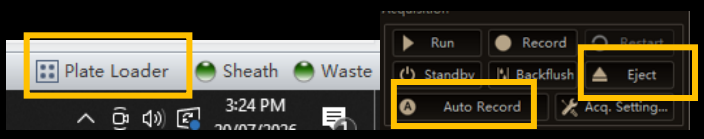
5/ Rotate sample injection switch to the left. The “P” will face forwards





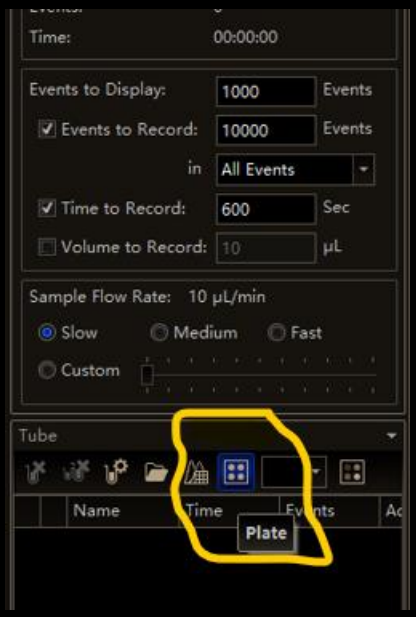
6/ Wait 5 seconds after power off before powering the Flex back on.

The following software changes will confirm the switchover:

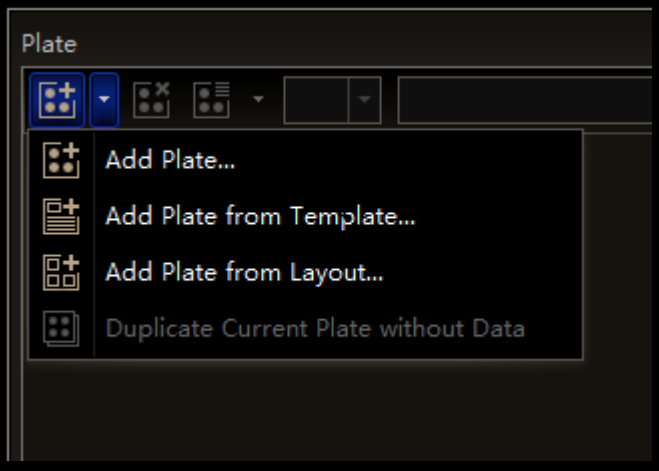


New experiment

1/ Click the plate button



2/ [Add plate], click the downward arrow on the first icon

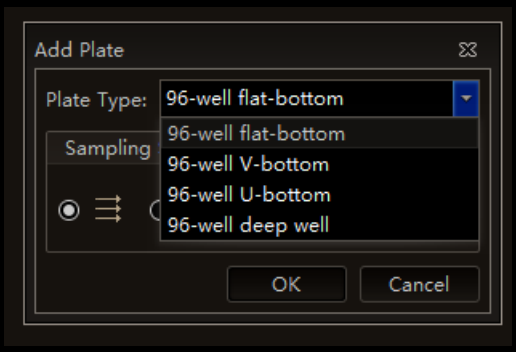


3/ Select plate type and sampling direction.

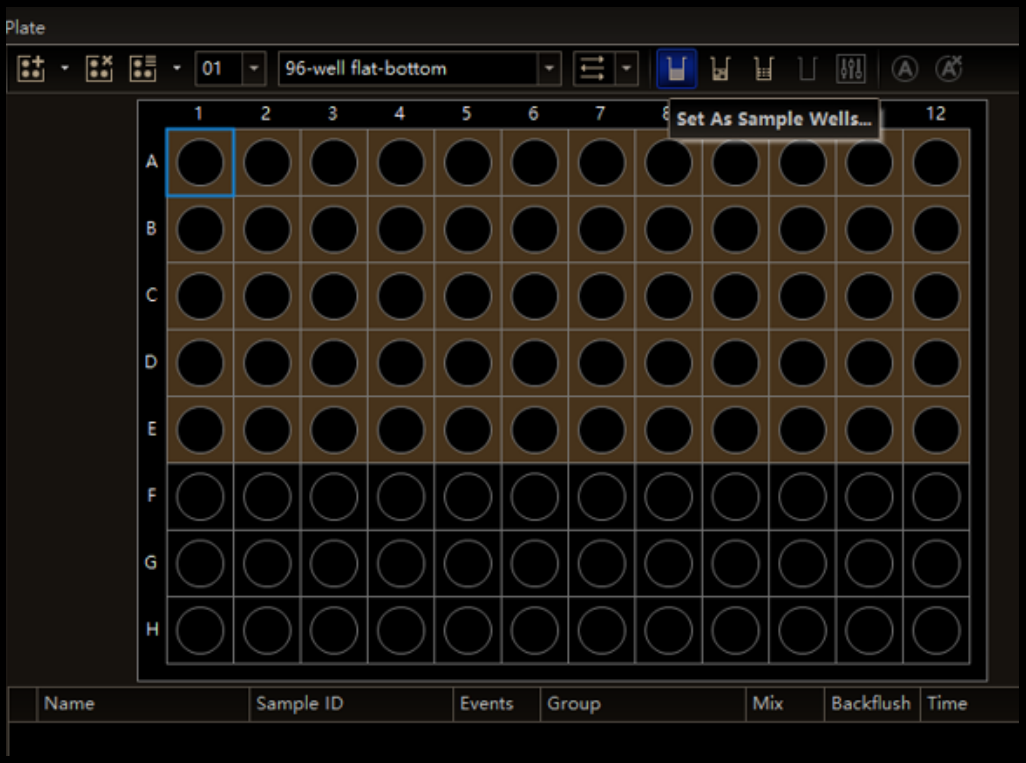
Calibrated for falcon plates and legendplex plate

/!\ other plate brands need to be calibrated before use. Check with facility staff.

/!\ deep well plates require an attachment. Check with facility staff beforehand

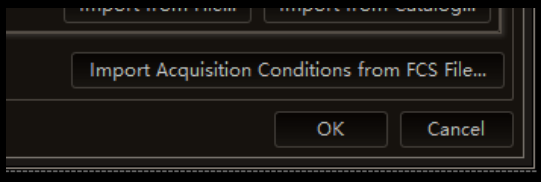


4/ A plate layout map will appear



5/

Gains not previously set	Select one well with a full stain sample and click [set as sample well]
Gains established	Click and deag to select wells containing sample > [set as sample well] Can import settings from an existing FCS file. Bottom right of [set as samples wells] window.



6/

A/ **Prefix**: label for each sample. Well ID (A1, A2) added to end of each sample Applies to all damples selected. If you want different names, need to perform multiple selections.

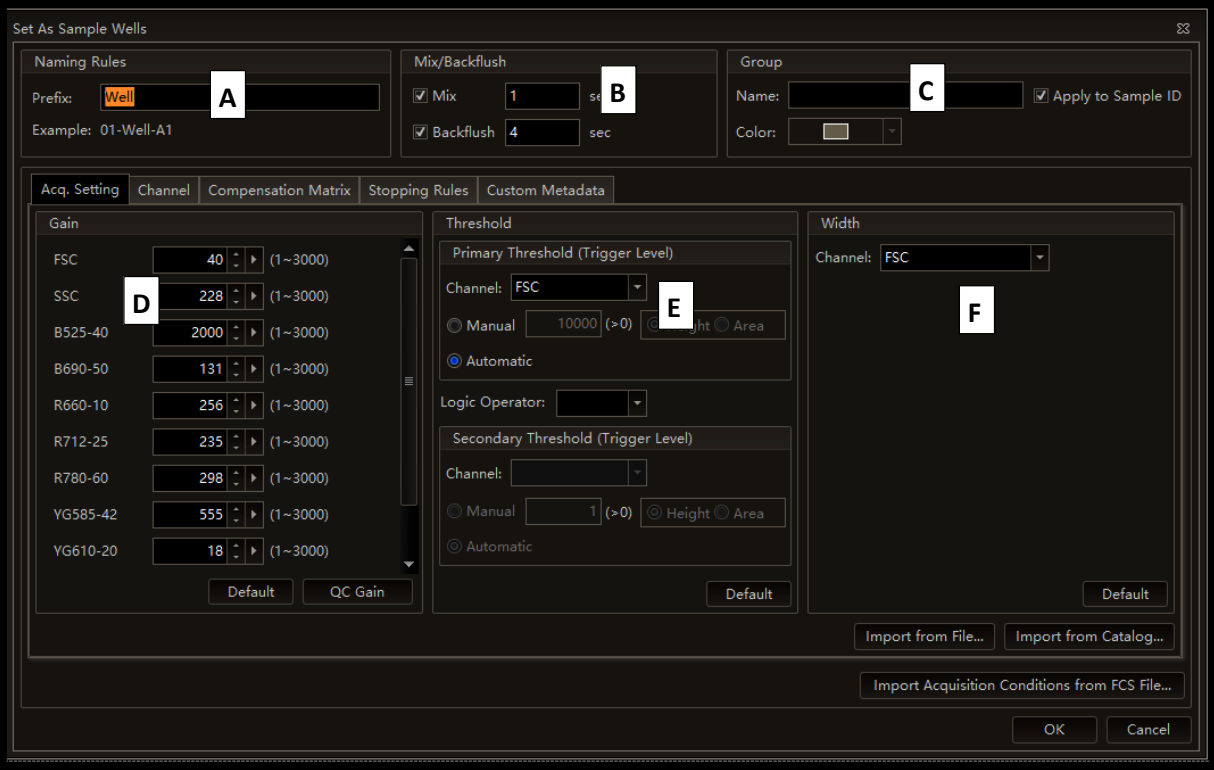
B/ **Mix** and **backflush** enable. 2 sec mix / 4 sec (5 for legendplex) backflush a good start.

C/ **Group** name doesn't do much. The **colour** will be applied to the wells on the plate layout window.

D/ **Adj. setting**. Set gains if known. If unknown set when running the sample.

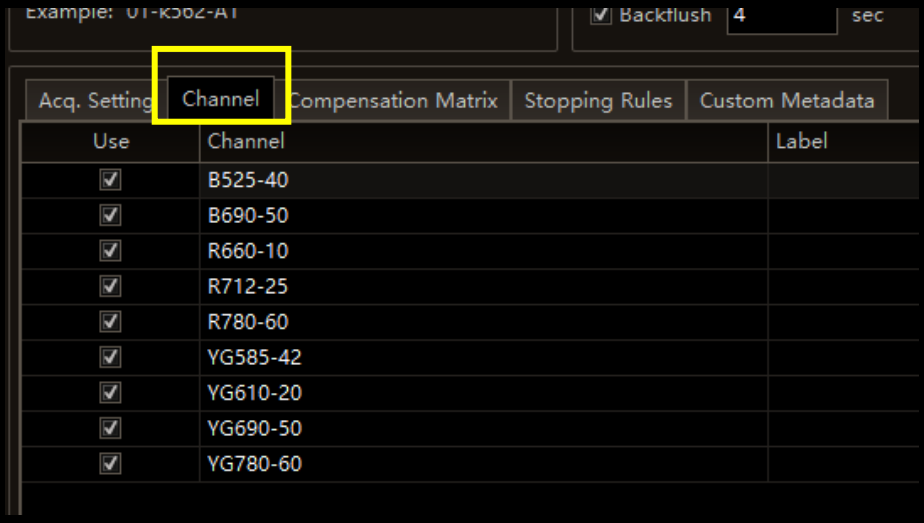
E/ Threshold. Can now use two parameters. Automatic/ FSC will be fine for most experiments. If using two channels, yuse the OR logic operator.

F/ Width. FSC default. For cell cycle, can use the DNA-stain channel.



7/ Select the [Channels] tab

Enable and label channels you will be using.



8/ select [stopping rules] tab

Set stopping criteria for event number, time, or volume

9/ If happy with set up, click OK in bottom right

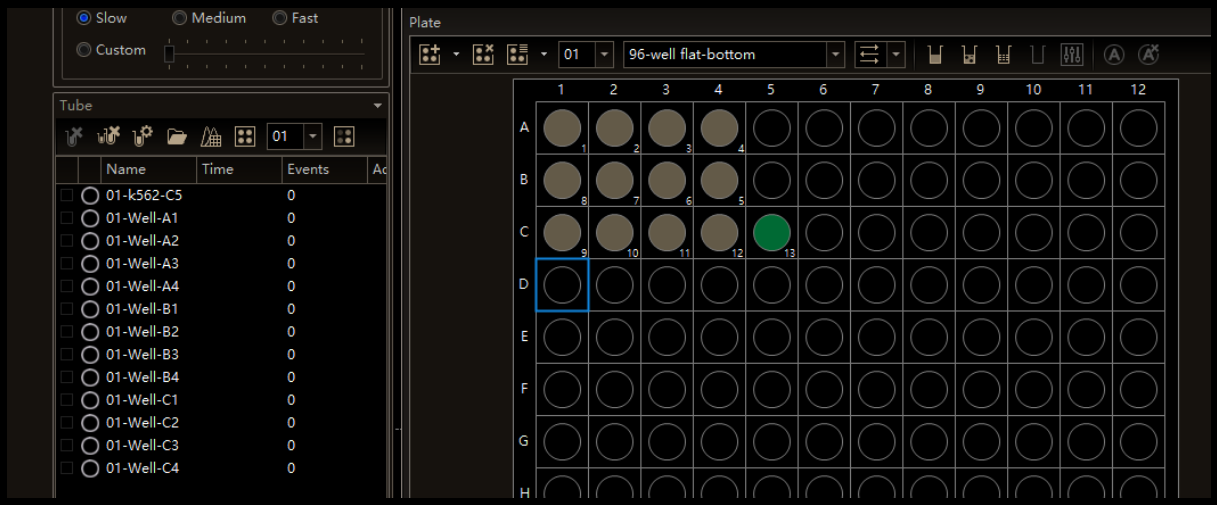
Can not get back to this screen. Will have to edit each option type separately.

10/

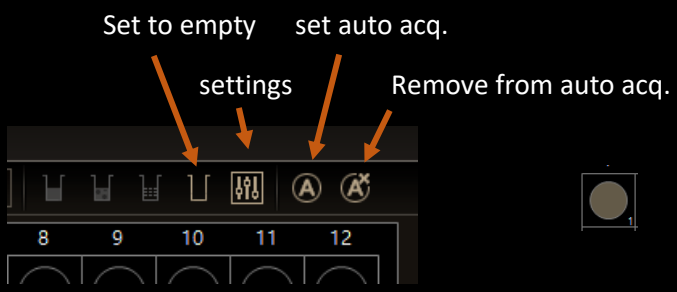
Samples will be generated.

They will be listed in the left [Tube] area.

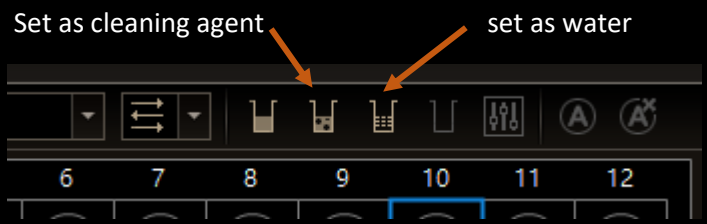
The plate layout will be coloured according on the wells previously selected.



The number in the bottom right of each well denotes the running order. You can remove the numbe to prevent from auto running.



You can set water and cleaning wells within the plate if required



11/ Set plots, same method as in tube mode.

12/ Press [eject] to open plate tray

13/ Load plate, ensure in place

14/ Click [Load ^] to close plate tray

15/ Select well to run (in left hand side list or plate window)

16/ Run sample

[Run] will run the selected well (no stopping criteria)

[Record] will record data for the selected well and stop baed on criteria

[Auto Record] will automatically record data for all the wells slated for auto run (if there is a number in the bottom right of the well in the plate window).

17/ (no gains established)

After establishing gains, for remaining wells, set them as sample wells (step 5).

17/ (gains known)

Manually acquire or auto record remaining sample-wells

Plate clean program

1/ There is a plate for the daily clean under the flex: front-right.

Fill 3 wells with facsClean

Fill 3 wells with water

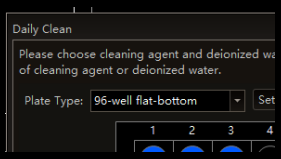
2/ Ensure “keep clear” area is clear.

3/ [Cytometer] > [daily clean]

Plate tray will open

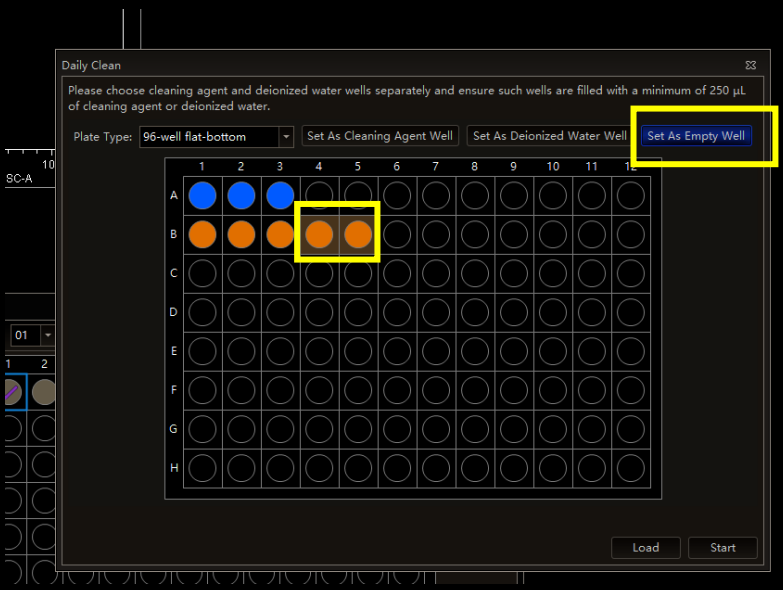
4/ Load plate on plate-tray.

5/ Select correct plate type.



6/ remove two water wells from plate layout

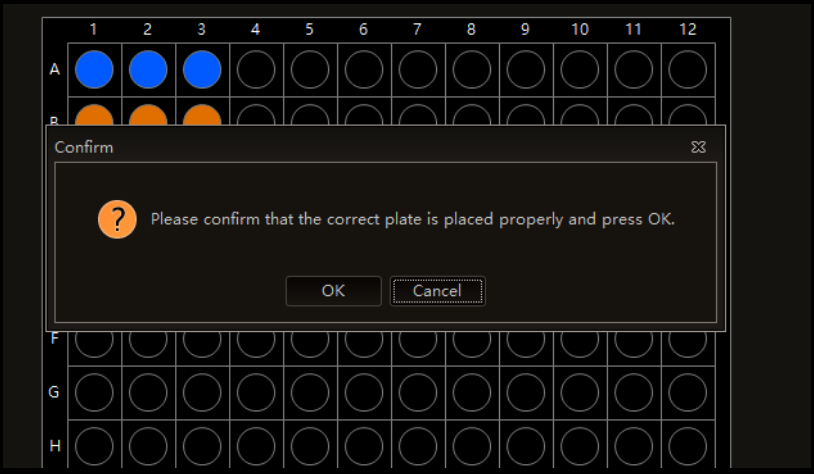
Select last two water wells, click [Set as empty well]



7/ Close plate tray by clicking the [Load] button

8/ Click [start].

If correct plate installed, click [OK]



9/ When clean finished, eject plate-tray and remove plate.