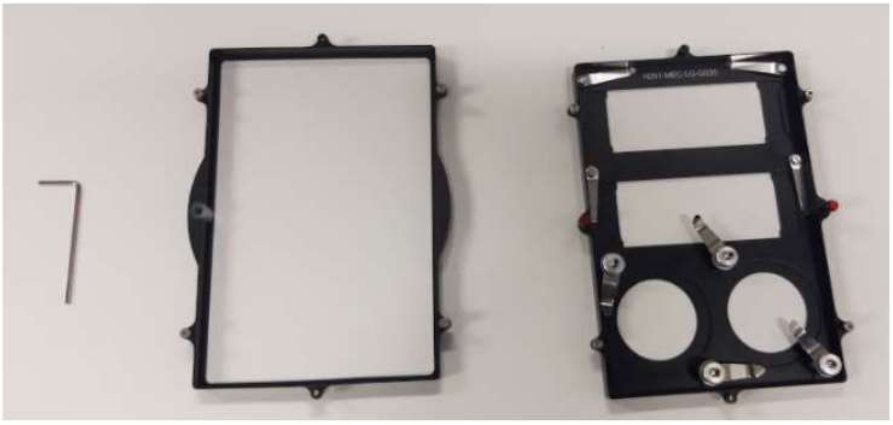
# Leica Stellaris 8 FALCON quick manual

Version 1.1

# Start-up

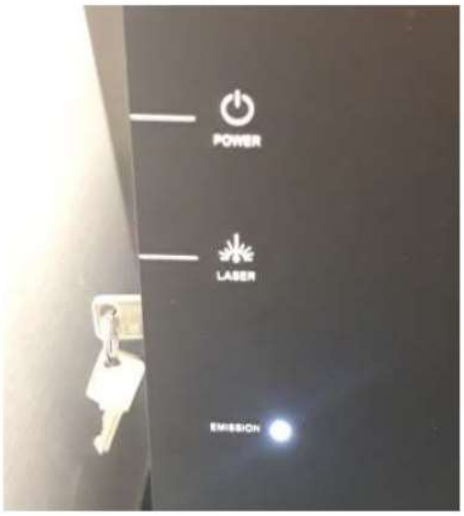
1. Check which stage adapter is in place. If you need to change the stage adapter, do it **before** you start the microscope.



The adapter on the left is meant for 96/24 wellplates and 3D-printed sample holders.

The adapter on the right is meant for regular objective glasses and 35 mm dishes. If you are working with incubation, remember to pluck the holes on the adapter using spare objective glasses / dishes found next to the microscope.

2. Turn on the microscope and the laser by pressing the two buttons on the large black box left of the microscope. Also turn on the fluorescent lamp.




You cannot see the switches, but you can feel the above the key. You do not need to touch the key unless it is not in the position shown here.

3. Start the computer and login with your local credentials.

4. Start the Las X software.

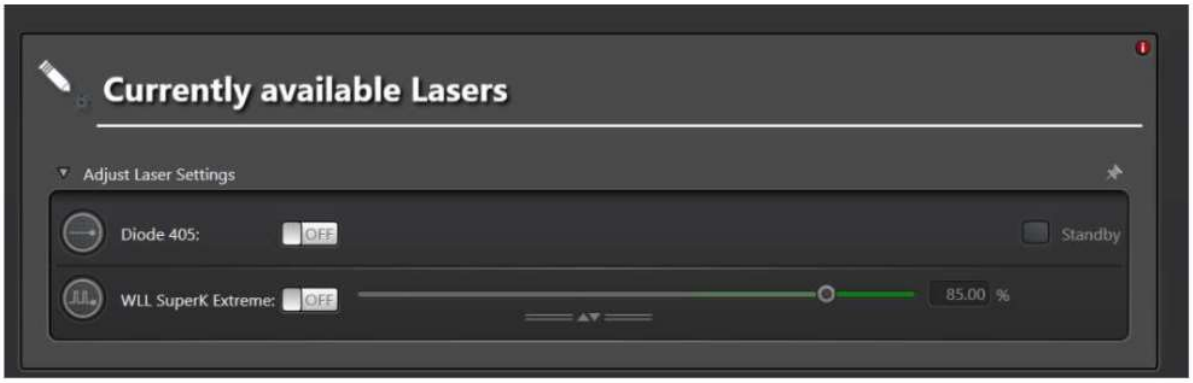

5. Chose the appropriate configuration. For the time being this is ‘machine’.


6. Once the software had fully started, you can put in your sample.

7. Got configuration menu (see below) and then chose laser configuration (see right)



This is where we turn on the lasers we need.




If you have DAPI / Hoechst in your sample, turn on the Diode 405 laser. Then turn on the white light laser (WLL). WLL will provide all other laser lines.

8. Now get back to the Acquisition side to set up the excitation / detection paradigm.



If you have any saved settings, you can load them from the button (A).

If you need to make a new scheme you can add new sequences form the (F) button. Then type the colour you have in your sample in the search window (B) and use mouse cursor to drag the desired dye from the dye bar (C) to the location of the desired detector (D) or (E). Adjust detection ranges if necessary.

Stellaris has three types of detectors, **HyD S, HyD X** and **HyD X**.

**HyD S** Not quite as sensitive for FLIM or photon counting as HyD X, but tolerate more light. Use these for bright channels and when making a coverslip correction.

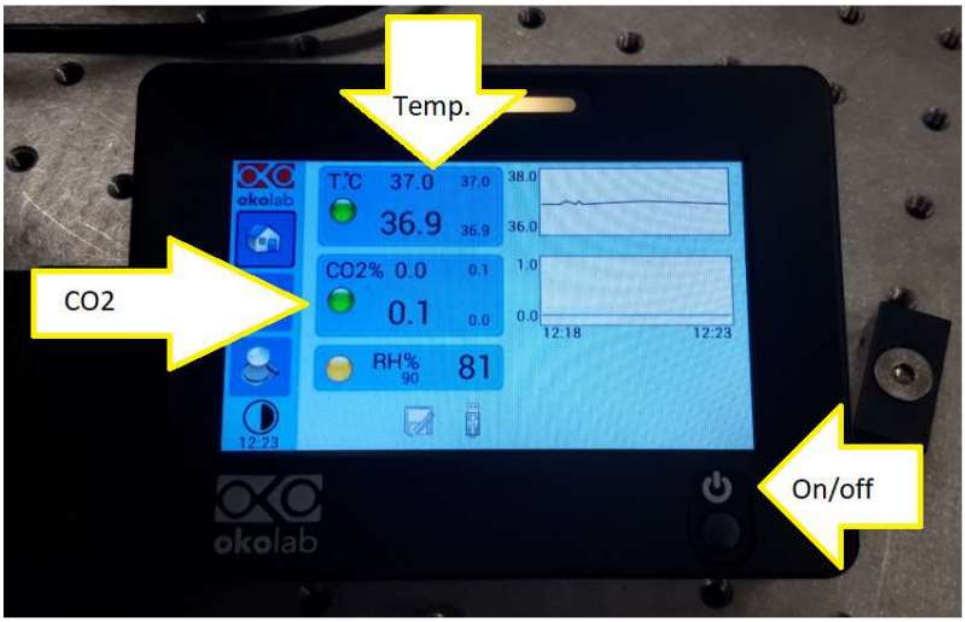
**HyD X** Use these detectors for dim channels / for photon counting applications / when doing FLIM. NEVER USE **HyD X** FOR COVERSLIP CORRECTION.

**HyD R** This detector is strictly meant for far red dyes and should be used with them. NEVER USE **HyD R** FOR COVERSLIP CORRECTION.

You can use widget (G) to visualize the excitation / emission spectrum of your chosen dyes to better design your excitation / detection paradigm.

## Heating and $CO_2$

The environmental controller is on the floatation table, next to the microscope. To turn on the heating press the on/off button. Adjust the temperature to either 37 C (red) or 29C (orange) depending on your needs or the colour of the next booking with 24 hours. Do not turn on the CO2 unless you need it yourself. 5% CO2 should be sufficient for most samples.




To turn off the heating, press the off button down for about five seconds. To turn off the heating, press the on/off button for about two seconds and then confirming the shutdown from the screen.

# Shutdown and preparing the microscope for the next user

1. Save your data.
2. Check who is coming next to the microscope and what lasers they need

3. Check from IRIS if the next user is coming in two hours or less.

   If the next user is coming in 2 hours or less, use protocol A, else use protocol B

A. Turn off the 405 laser from the laser configuration menu if you used it and the next user does not need it.
   Shut down the software.
   Remove your sample and clean the objectives.
   Log off from the computer.

   Check from IRIS if the next user within 24 hours needs the microscope heated. If you used heating, leave the heating on, adjusting temperature if needed. If you did not use heating, turn the heating on, adjusting temperature if needed.

   If you used $CO_2$ turn it off by setting the gas concentration to 0% from the controller and by closing the valve on the wall.

   disinfect all the surfaces you touched.

B. Turn off both the lasers from the laser configuration menu.
   Shut down the software.
   Remove your sample and clean the objectives.

   Shut down the microscope.
   Shut down the fluorescent lamp.

   Check from IRIS if the next user within 24 hours needs the microscope

heated. If you used heating, leave the heating on, adjusting temperature if needed. If you did not use heating, turn the heating on, adjusting temperature if needed.

Shut the computer down.

If you used $CO_2$ turn it off by setting the gas concentration to 0% from the controller and by closing the valve on the wall.

Disinfect all the surfaces you touched