

Instruction SHORT LSM880 2024-03-24.docx 2024-03-24

LSM 880 Quick Instructions

Switch on

- Make sure room temperature is 22-24°C.
- Turn on air condition
- In the main switch box turn on the following switches **1 → 2 → 3 (do not touch the key)**
- Turn on the computer and log in
User name: **multilabs**
Password: **123456**

Position	Objective lens	Magnification	NA	coverslip	Working distance	Immersion	Parfocal length
2	Plan Achromat	x10	0.45	0.17 mm	2 mm	Air	45 mm
3	Plan Achromat	x20	0.8	0.17 mm	0.55 mm	Air	45 mm
4 (bombila)	W Plan Achromat	x20	1.0	0	1.8 mm	Water dipping	75 mm
1+ adaptor	W Plan Achromat	x40	1.0	0	2.5 mm	Water dipping	45 mm
1+ adaptor	Plan Achromat	x63	1.4	0.17	0.19 mm	Oil	45 mm

Software and acquisition

- Log into **BookitLab** and activate your reservation to start **Black Zen 2.3**
- To acquire images click **Start System**
- In the acquisition tab → Laser tab
Switch on the lasers, (The Argon laser should be turned on by Standby → ON. Please wait 5' minutes until the laser stabilizes, its status should be **optimal** in the properties tab).
- **Sample mounting and viewing**
Insert the slide or dish in the holder.
To change objective turn the black wheel to point up to release the objective turret. Turn manually to the required objective, lower the objective by pointing the black wheel in the objective turret down, (if an objective needs to be replaced please ask the microscopy staff).
In the software go to Locate tab.
On the microscope's right side choose VIS
- **Transmitted light**
Click the transmitted light lamp icon, click ON and set desired brightness (or via the microscope).
Set the filter turret position to DIC and the condenser below the stage to II or III according to the objective lens (see objective lens info on touchpad).
Set up Kohler illumination.
- **Reflected light (fluorescence)**
Open fluorescence shutter
Set the filter turret to the desired position
- **Image Acquisition**
On the microscope's right side choose LSM
In Zen, **Acquisition** tab:
Load a saved configuration from the **Experiment Manager** or open an image and below the image click **Reuse**
Select **Show all tools** and open the following tabs:

Light Path, Channels, Acquisition Mode

With one track checked, in fast settings (512x512, speed 8, no averaging) click **Continuous** to focus.

Optimize each channel separately by checking **Range Indicator** and setting pinhole (1 Airy unit) detector gain, laser power, offset, digital gain.

Check all channels and set acquisition parameters: zoom, pixel number, scanning speed, averaging.

After setting zoom press "optimum" so that the system will calculate number of pixels according to the optimal Nyquist number for the given objective. If scanning time is too high you can consider saving time by:

- lowering pixel dwell time.
- setting scanning direction to meander instead of comb
- decreasing number of averaging iterations
- using frame instead of line mode

Press **Snap** to acquire image

For multidimensional images check the desired dimension (Z, time, tile) and after setting the relevant parameters click **Start Experiment**

See instructions booklet for more details

- Saving images

Save image as CZI filetype in **D:\LSM 880 USERS DATA\PI name\user name\mm-yyyy**

- Data backup

Data are copied automatically every night, if computer is on and connected to the internet.

PLEASE do not use any type of USB flash drive to retrieve your data.

There are computers with active scanning anti-virus software at the Computerized Analysis Room, connected to the servers.

Ending session

Before you finish please make sure the next user is coming.

If yes:

- Close the ZEN software
- **Log off your reservation at BookItLab**
- clean oil immersion objectives with lens tissue dipped in petroleum ether, twice
- clean water dipping objectives by dipping in DDW (e.g., in 35mm dish)
- Leave nosepiece at 10x objective lens
- Center the stage
- Leave surfaces clean

If no user is coming in the next two hours, also, **in this order:**

- Turn off the lasers in the software
- Close the ZEN software
- Log off your reservation in BookItLab
- Switch off **3 → 2** in the switch box
- **Wait until the Lasos fan shuts off and only then switch off → 1**
- Cover microscope
- Turn off air condition

Do not turn off computer unless you are the last user before the weekend.

Please do not use any type of USB drive to copy your data.