

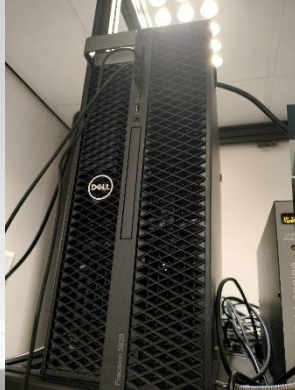
A photograph of a Nikon Spinning Disc Confocal microscope setup in a laboratory. The microscope is mounted on a black metal frame with a white base. To the left, there is a control console with a joystick and buttons. In the background, a computer tower and other lab equipment are visible on a shelf. The setup is on a perforated metal table. A white banner with black text is overlaid on the top of the image.

Nikon Spinning Disc Confocal-User Guide

Kavli Nanolab Imaging Center

Turn ON- Hardware

6. PC



2. Nikon Controller



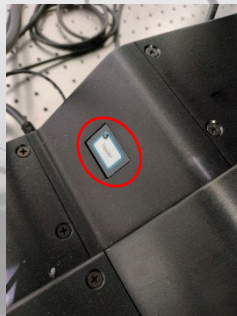
5. FRAP Ctrl



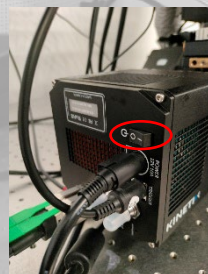
On table,
behind



4. SD unit



3. Camera



I. main switch



Don't forget to turn off the chamber light



! Make sure the objective is far away (<math><500</math>) before changing holders

Available stage holders

Multi-well plate



Single 35mm dish



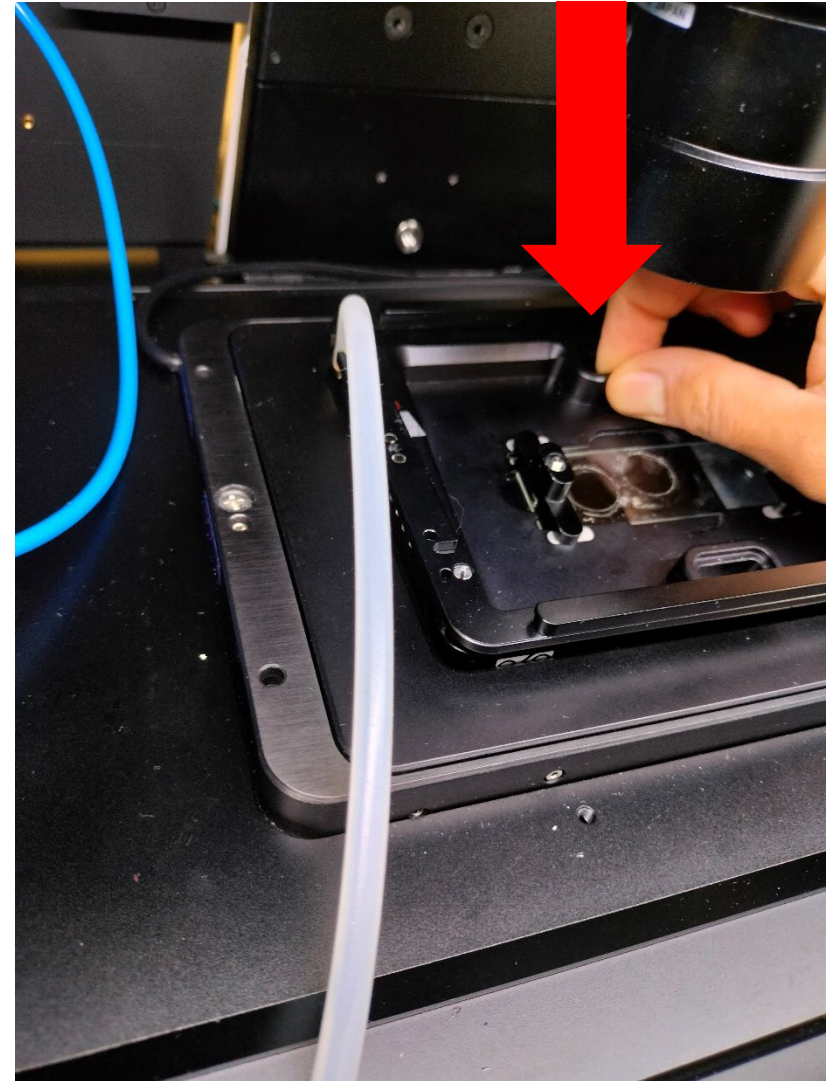
Slides



double 35mm dish

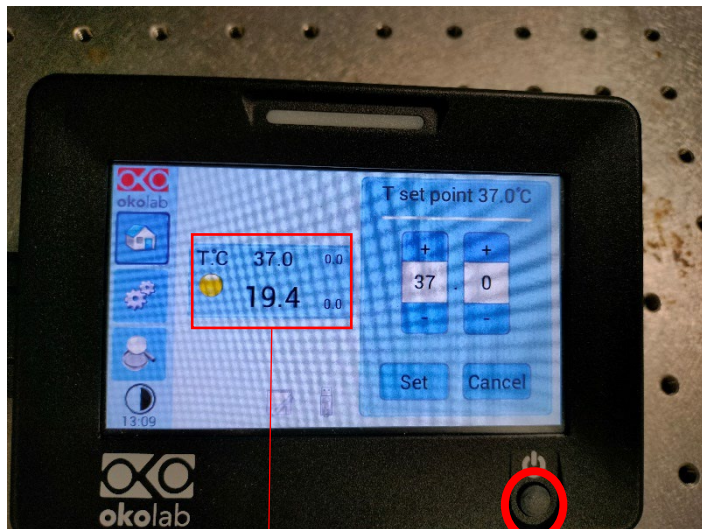


Place in/out



Environmental control

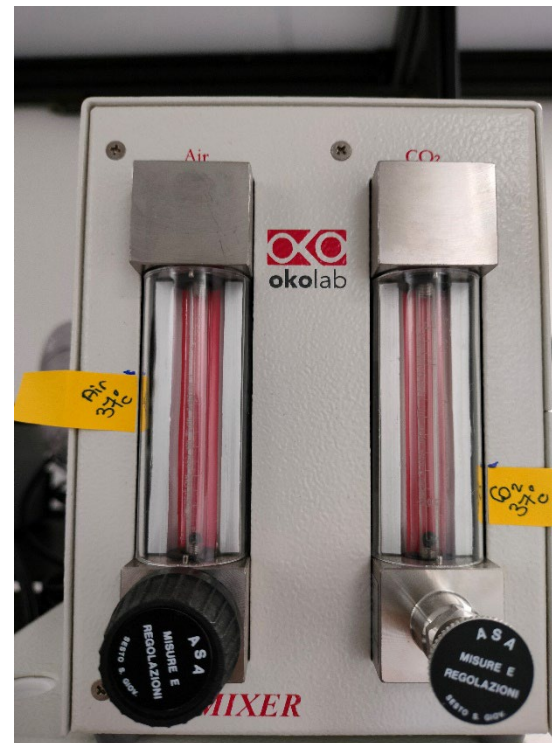
Temp Control



1, Turn on the controller

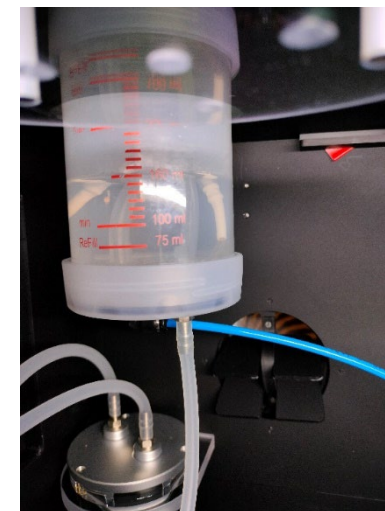
2, click to set desired temperature

CO₂ Control

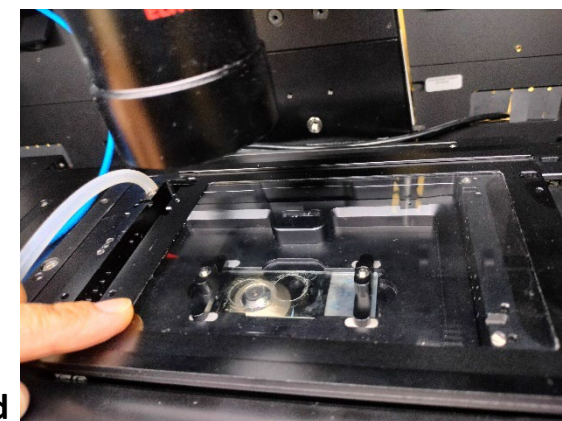


1, Open the valves until levels are stabilized to 5% setpoint

2, Check the level in humidifier is sufficient. If needed, fill with DI water



3, Close the lid

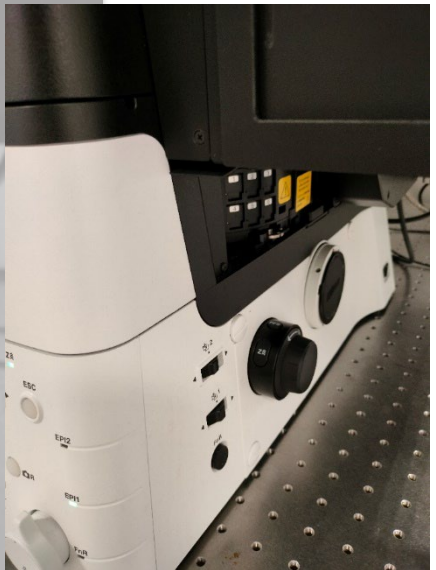
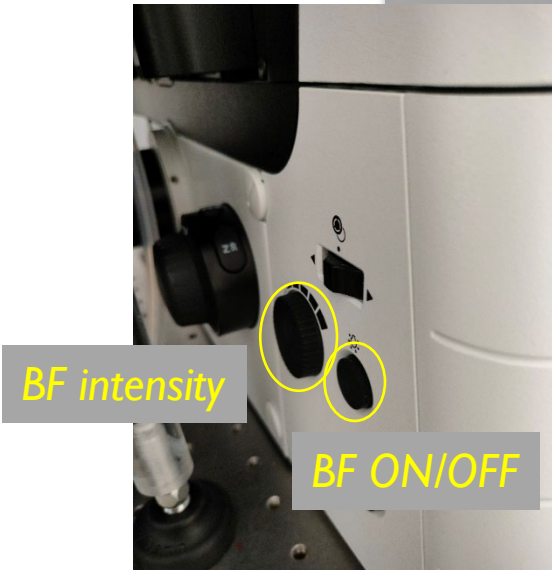


Nikon Ti2- how to



!Note- its better to operate all **Eyepath** channels (BF/Flu) from the software

When PFS finds the surface, you will hear a “beeping” sound. You can then activate (turns green) it to keep Z position



No need to change filter cubes manually- operate through software!

If you used **ESC-** to release, **press and hold** to avoid jumping back to position

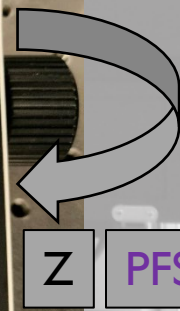
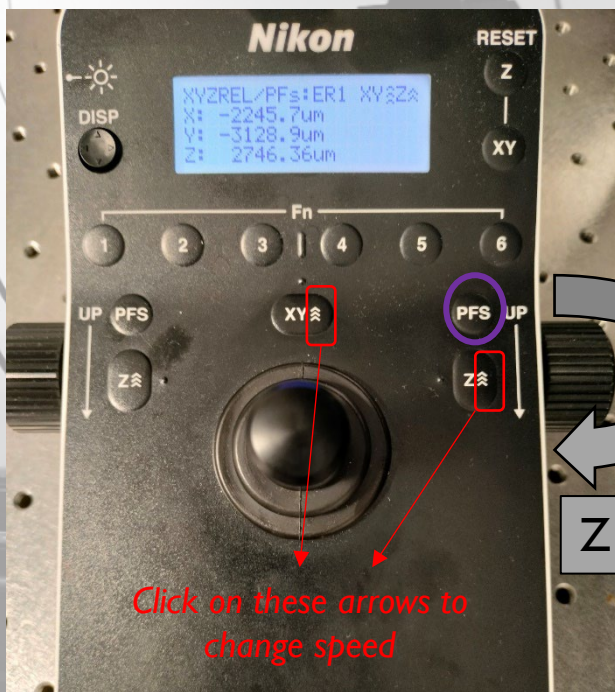
Nikon Ti2- Joystick



1, objective and light path

2, Stage positions, PFS

Press from the side to shift info views:



Click on these arrows to change speed

Note- optional objectives exist: 20X/40X air; 60X Water, 100X oil

Objectives (from Nikon):

Objective type	Mag/NA	Medium	Contrast	WB (mm)
Plan Achrom	10x / 0.25	Air	Fluo, Ph1	10.5
Plan Apo λ	20x / 0.75	Mimm	Fluo, DIC N2	0.35 (Oil) 0.34 (Gly) 0.33 (W)
Plan Apo λ	60x / 1.4	Oil	Fluo, DIC, Phase	0.13
CFI SR HP Apo λ S	100x / 1.35	Silicon	Fluo, DIC, Phase	0.3

Nis Elements

Your Work Interface looks like this:

Acquisition controls

Phase	Interval	Duration	Loops
#1	1 sec	999	1

This is your design of experiment:
Time/Z/XY/channels
Save each experiment in your own folder and name it properly

Set Iris

Format: No Binning
Bit Depth: 16-bit
Auto Exposure: 400 ms
Temperature: 0.0 °C

Cofirst pod
365 nm 440 nm 488 nm 514 nm
561 nm 640 nm 730 nm
365 nm 67.62 [%]
488 nm 80.00 [%]
561 nm 61.07 [%]
640 nm 72.55 [%]
730 nm 100.00 [%]
Temperature: 22°C Supply: 450mA, 20W
Cofirst Configure...

DIA
0.0 100.0 23.8

Filters
TurnOff

X-Light Pad
EM Wheel
Illumination Iris 100.0 [%]
Emission Iris 50.0 [%]

***Set Iris Macro Panel:**

Click this once per objective, to set emission Iris correctly

Image controls

Over saturation

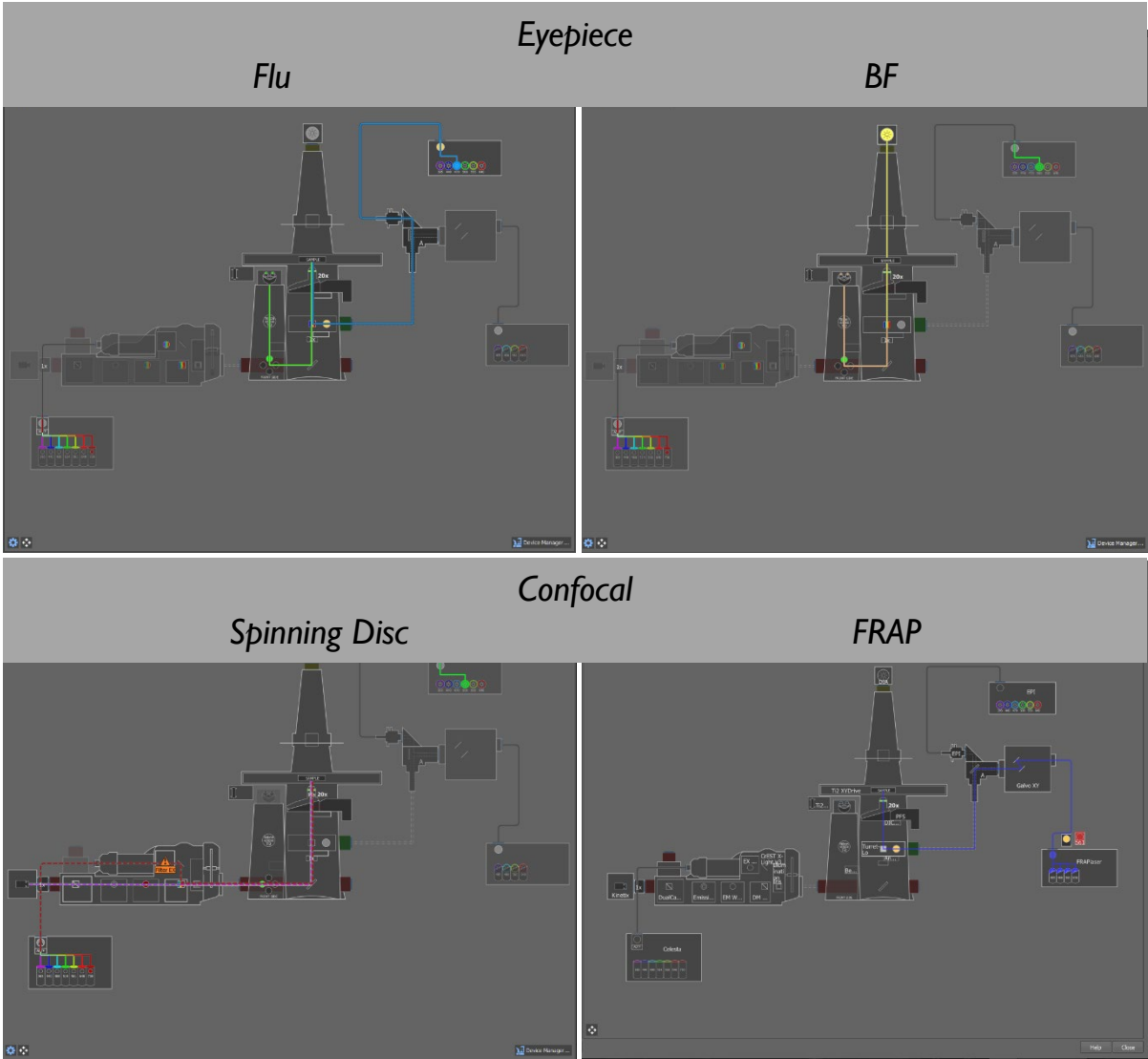
Histogram

Opened Images

Here you can toggle between acquired images.
Ctrl "i" shows image information

Nis Elements

Light Path defines your interface, the illumination source and detection device:



Nis Elements- Eyepiece

You can shift between BF and Epi-Flu

Click on each of the **presets** to activate the suitable illumination-detection settings



To see BF, make sure the shutter is on "O" (open) position and not "C" (closed).

Acquisition x

Live Capture ND Acquire Large Image Save Save As... Open

Eye - Eye - EPI Spinning Disk Lightpath

Brightfield DAPI GFP TxRed Cy5 Add

Kinetix Pad

Format: No Binning

Bit Depth: 16-bit

Auto Exposure: 200 ms

ROI: ROI Size

Temperature: 0.0 °C

Ti2 Pad

Nosepiece: 100x 10x 20x 60x

Zoom: 1x

PFS: Glass Dichroic Offset

PFS: 78.49

Spectra Pad

395 nm 440 nm 470 nm 508 nm 555 nm 640 nm

1: 395 nm 57 [%]

2: 440 nm 43 [%]

3: 470 nm 52 [%]

4: 508 nm 35 [%]

5: 555 nm 42 [%]

6: 640 nm 9 [%]

DIA: 0.0 100.0 28.7

Shutters: FL-Lo

Filters: Turret-Lo

Configure...

EPI Configure...

Acquisition x

Live Capture ND Acquire Large Image Save Save As... Open

Eye - Eye - DIA Eye - EPI Spinning Disk Spinning Disk - HS Lightpath

Brightfield DAPI GFP TxRed Cy5 Add

Kinetix Pad

Format: No Binning

Bit Depth: 12-bit

Auto Exposure: 500 ms

ROI: ROI Size

Temperature: 0.0 °C

Ti2 Pad

Nosepiece: 100x 10x 20x 60x

Zoom: 1x

PFS: Glass Dichroic Offset

PFS: 78.49

Spectra Pad

395 nm 440 nm 470 nm 508 nm 555 nm 640 nm

1: 395 nm 18 [%]

2: 440 nm 13 [%]

3: 470 nm 12 [%]

4: 508 nm 24 [%]

5: 555 nm 27 [%]

6: 640 nm 9 [%]

DIA: 0.0 100.0 86.4

Shutters: FL-Lo

Filters: Turret-Lo

Configure...

EPI Configure...

You can see the objectives here. **!Note!** only choose from Ti2 body **before mounting sample!** **Do not click to avoid auto switch!**

Here you can see full obj info

Nosepiece position	Objective name	Z-Step (Auto Focus)	Z-Step (Slices)	Working distance
1	SR HP Plan Apo Lambda S 100	0.60	0.30	300.00
2	Uncalibrated			
3	Plan 10x	16.00	8.00	10500.00
4	Plan Fluor 20x MImm DIC N2	1.80	0.90	350.00
5	Plan Apo IR 60x WI DIC N2	0.60	0.30	180.00
6	Uncalibrated			

Define optional objectives... OK Cancel

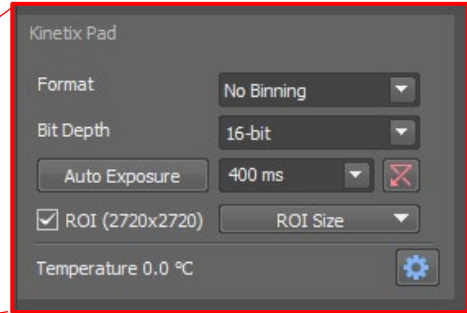
Nis Elements- SD

You can shift between BF (transmitted) and Confocal-Flu

Click on each of the **presets** to activate the suitable illumination-detection settings

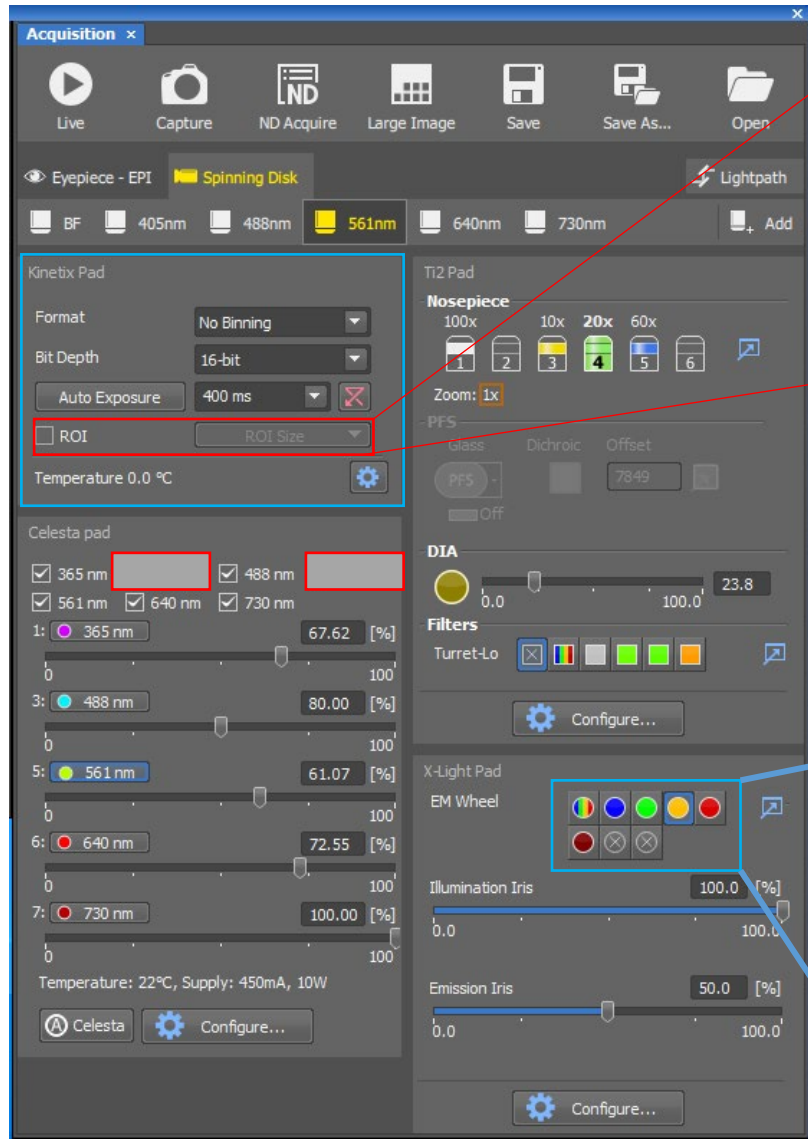
Camera settings.
Recommended*:
No binning
16bit
Minimal exposure time

Missing filters.



ROI size:
If unchecked, you see full FOV-
more than illuminated area
Crop to 27mm or lower, or
define your own ROI on image

Emission filters- matching excitation.
for fast imaging- use multiband cube
*First check controls for channel leakage!



*Note that imaging lasers are strong and can bleach fast sensitive samples. If this is an issue:

Try to bin (sCMOS pixel size is smaller than EMCCD), lower laser as possible and work in low signal regime, with post-enhancements (Huygens)

Nis Elements- ND acquisition

Image large area- better use PFS!!

The screenshot shows the 'ND Acquisition' dialog box. At the top, the 'Experiment' is set to 'ND Acquisition'. Below this are fields for 'T:', 'L:', and 'λ:'. A 'Save to File' checkbox is checked, with the 'Path' set to 'C:\Data\Michal\test capacity PC memory' and the 'Filename' set to 'timelapse_405no delay 10h003.nd2'. A 'Record Data...' button is visible. There are also options for 'Custom Metadata', 'Order of Experiment', and 'Timing...'. A row of icons includes 'Time', 'XY', 'Z', 'Large Image', and 'λ', with 'Time', 'Large Image', and 'λ' checked. The 'Scan Area' section has radio buttons for '2 x 2 fields', '6,0 x 6,0 mm', and 'Pattern'. The 'Stitching' section shows 'Overlap' at 10% and 'Stitching via' set to 'Blending'. At the bottom, there are checkboxes for 'Close Active Shutter during Stage Movement' and 'Use PFS', with the latter highlighted by a yellow box. A 'Run now' button is at the bottom right.

The screenshot shows the 'Time Schedule' dialog box. At the top, there are icons for 'Time', 'XY', 'Z', 'Large Image', and 'λ', with 'Time' and 'λ' checked. Below is a table with columns 'Phase', 'Interval', 'Duration', and 'Loops'. The first row is selected and highlighted in blue, showing '#1', '1 sec', '???' (with a dropdown arrow), and '1'. Below the table are checkboxes for 'Close Active Shutter when idle', 'Perform Time Measurement (0 ROIs)', and 'Switch Transmitted Illuminator off when Idle (0.01 s)'. The 'Use PFS' checkbox is checked and highlighted with a yellow box. At the bottom, there are 'Events...' and 'Advanced >>' buttons, and a 'Run now' button.

Phase	Interval	Duration	Loops
<input checked="" type="checkbox"/> #1	1 sec	???	1
<input type="checkbox"/>			
<input type="checkbox"/>			
<input type="checkbox"/>			
<input type="checkbox"/>			
<input type="checkbox"/>			

Nis Elements- ND acquisition

Time experiment- use PFS!!

ND Acquisition x

Experiment: ND Acquisition

T:

M:

λ:

Save to File

Path: C:\Data\Michal\test capacity PC memory

Filename: timelapse_405no delay 10h003.nd2 ND2

Custom Metadata

Order of Experiment

Time XY Z Large Image λ

Time Schedule

Phase	Interval	Duration	Loops
<input checked="" type="checkbox"/> #1	1 sec	???	1
<input type="checkbox"/>			
<input type="checkbox"/>			
<input type="checkbox"/>			
<input type="checkbox"/>			

Close Active Shutter when idle Use PFS

Perform Time Measurement (0 ROIs)

Switch Transmitted Illuminator off when Idle (0.01 s)

1 time loop

Set wavelengths (taken from your acq parameters)

ND Acquisition x

Experiment: ND Acquisition

T:

M:

λ:

Save to File

Path: C:\Data\Michal\test capacity PC memory

Filename: timelapse_405no delay 10h003.nd2 ND2

Custom Metadata

Order of Experiment

Time XY Z Large Image λ

Setup

Opt. Conf.	Name	Comp. Color	T Pos.	Focus Off...
<input checked="" type="checkbox"/> Spin...:561nm	561nm		All	
<input checked="" type="checkbox"/> Spin...:488nm	488nm		All	0
<input checked="" type="checkbox"/> Spin...:405nm	405nm		All	0
<input type="checkbox"/>				

Close Active Shutter during Filter Change Use PFS Use Trig. Acq.

Use Ratio

1 time loop

Nis Elements- ND acquisition

Z stack

Absolute- Piezo

ND Acquisition x

Experiment: ND Acquisition

T:

M:

λ:

Save to File

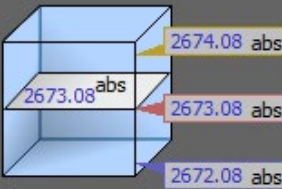
Path: C:\Data\Michal\test capacity PC memory

Filename: timelapse_405no delay 10h003.nd2 ND2

Custom Metadata

Order of Experiment

Time XY Z Large Image λ



Step: 0,3 μm ← 0,9 μm 8 Steps Range: 2.00 μm

Bottom: 2672.08 μm Top: 2674.08 μm

Z Device: Ti2 ZDrive

Relative Positions:
Top: +1.00 μm
Bottom: -1.00 μm

Close Active Shutter during Z Movement Direction: Bottom to Top Top to Bottom

Relative- you can use PFS and objective (Ti2) Zdrive

ND Acquisition x

Experiment: ND Acquisition

T:

M:

λ:

Save to File


Path: C:\Data\Michal\test capacity PC memory

Filename: timelapse_405no delay 10h003.nd2 ND2

Custom Metadata

Order of Experiment

Time XY Z Large Image λ



Step: 0,3 μm ← 0,9 μm 9 Steps Range: 2.00 μm

Range: 2 μm <-1.00, +1.00>

Z Device: Ti2 ZDrive

Relative Positions:
Top: +1.02 μm
Bottom: -0.98 μm

Close Active Shutter during Z Movement Direction: Bottom to Top Top to Bottom

Nis Elements- stage overview tools

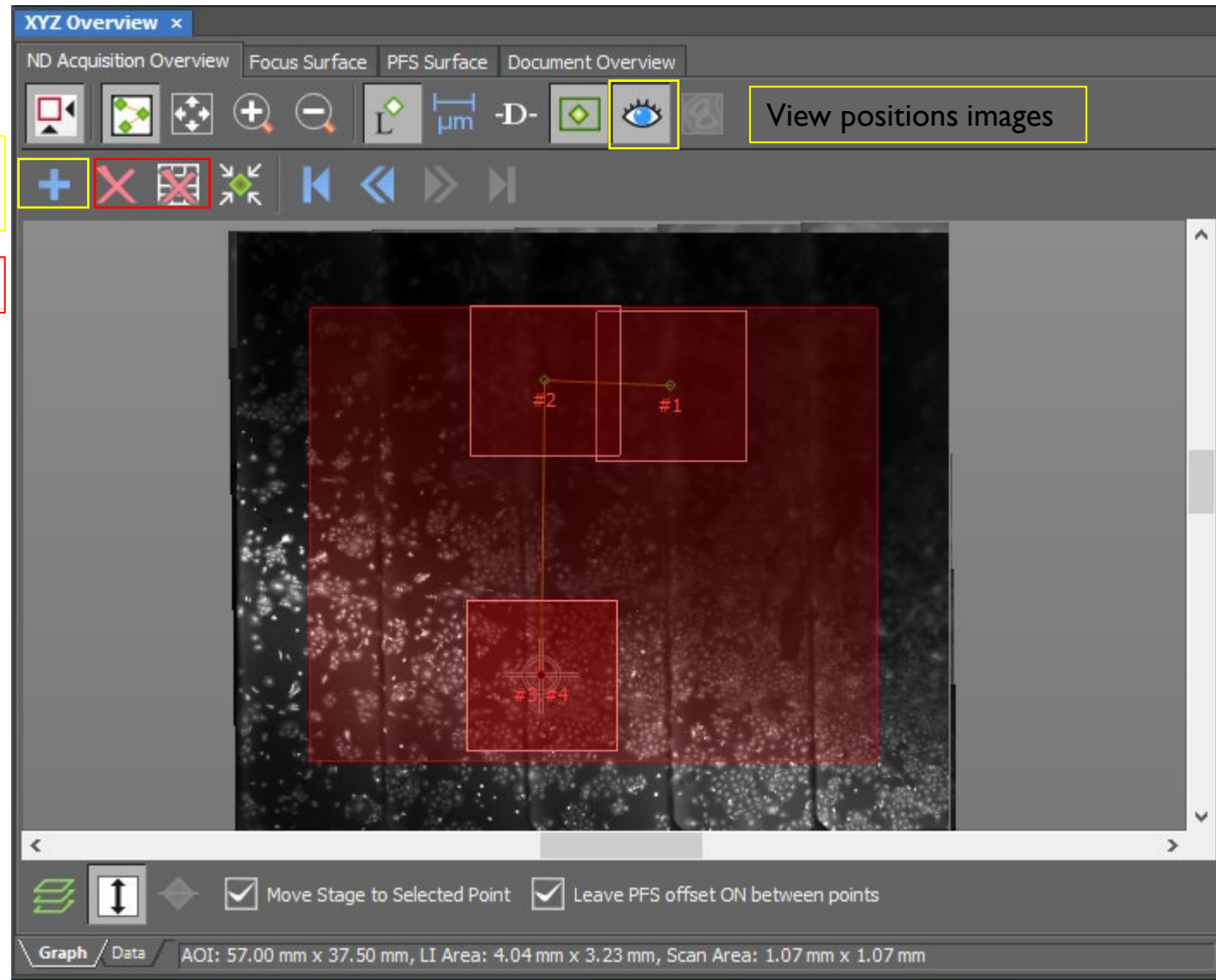
You can run fast preview scan or large image scan

Add new position
(also added to ND acq list)

Delete one point or all

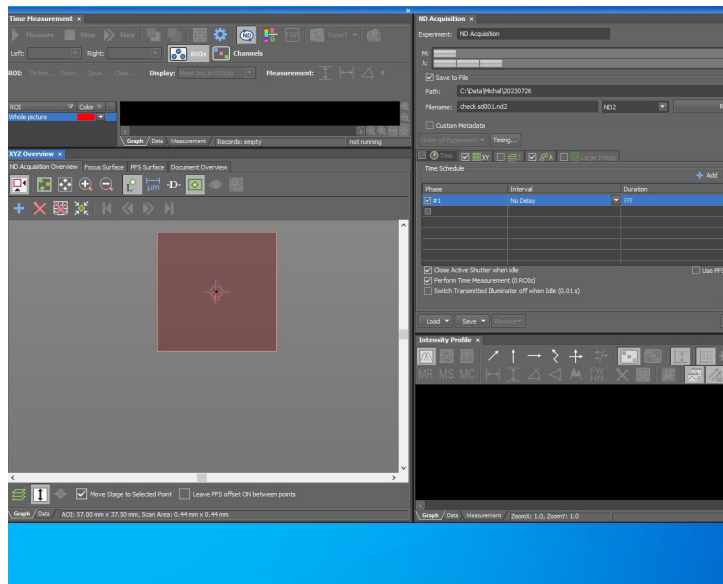
Doble click on position will move
the stage there

Right click will allow you to open
a dialog box in which you can
define large image scan.

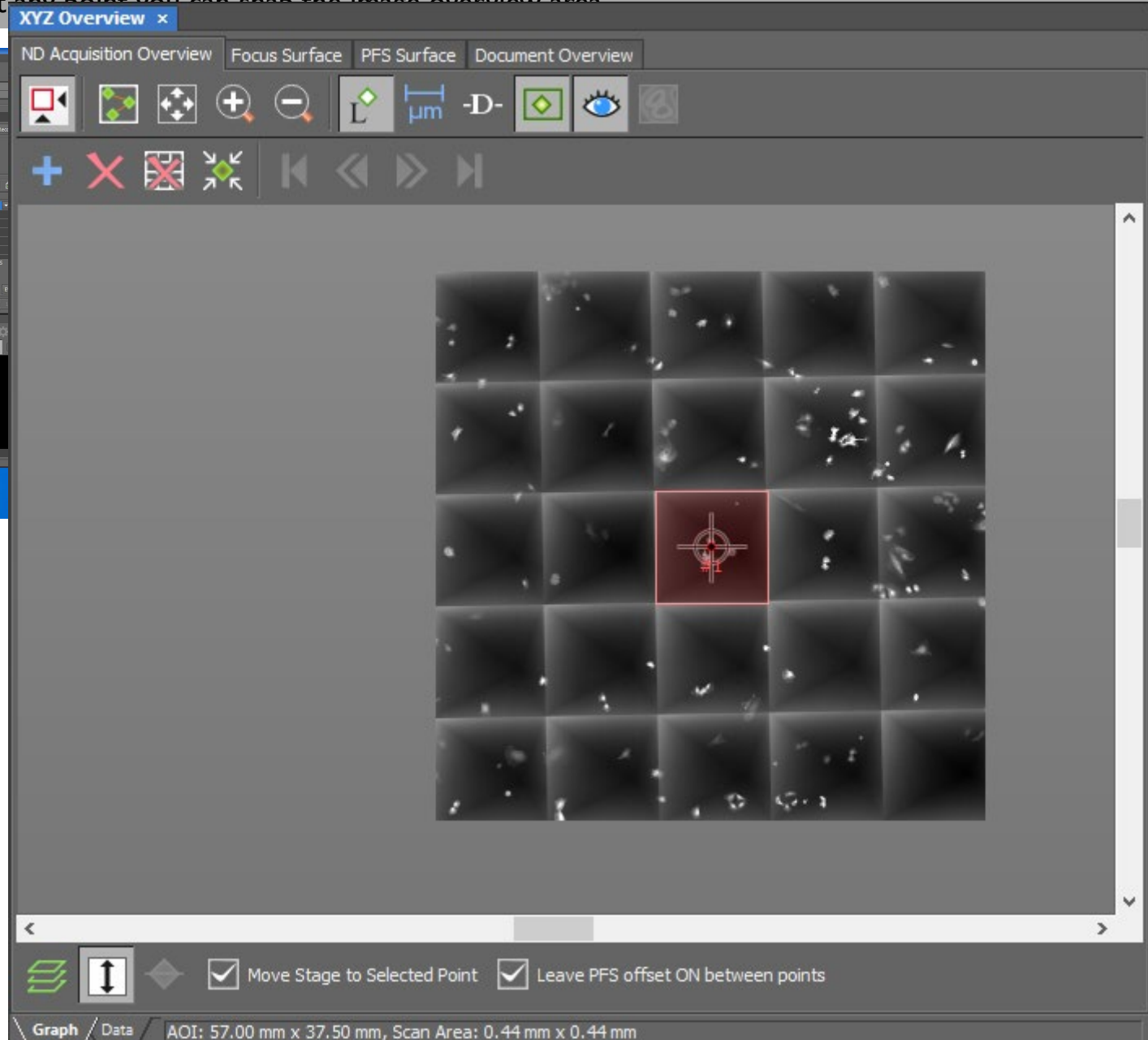


Nis Elements- stage overview tools

At any point you can snap the image overview area



Right click will allow you to open a dialog box in which you can snap the overview and save.

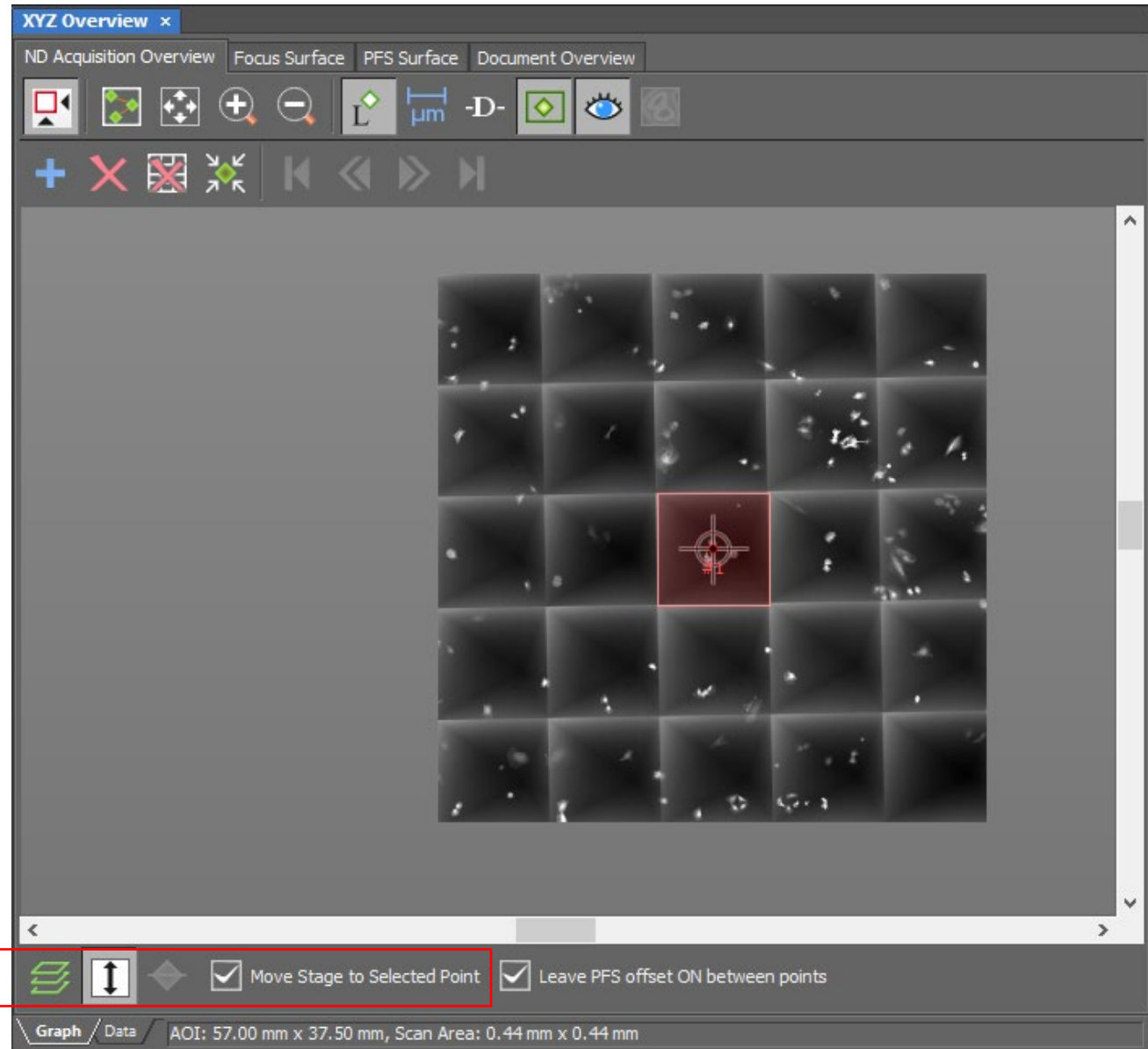


Nis Elements- stage overview tools

Fast Preview

Right click will allow you to open a dialog box in which you can define preview image to be scanned quickly (one channel, one plan, no stitching).

This preview can be further used as a map for navigation.

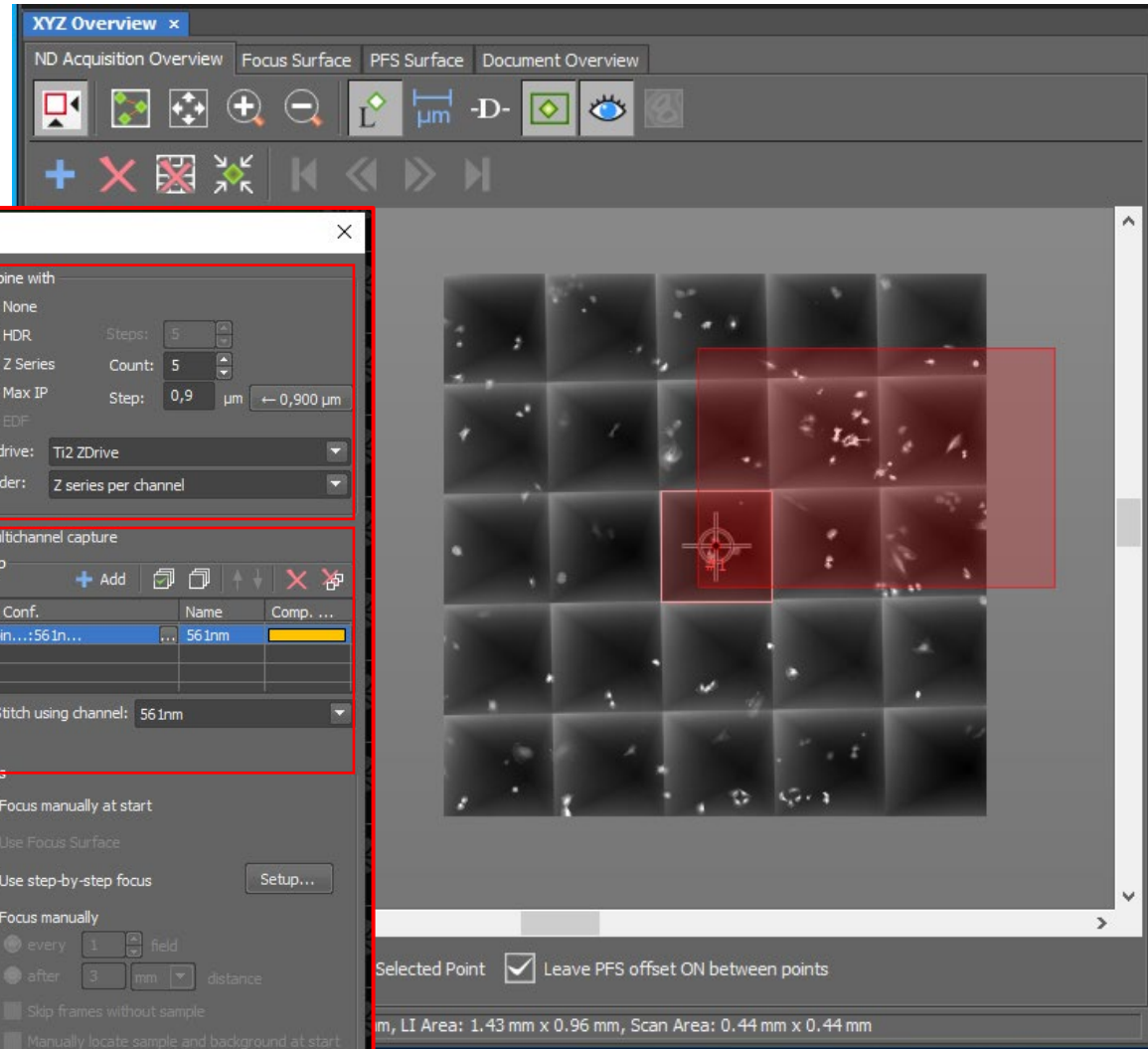


Double click on any area in the preview will move stage

Nis Elements- stage overview tools

Large image scan options

Right click will allow you to open a dialog box in which you can define area for large imager scan. It is similar to ND large image, but with more advanced built in options:



Scan Large Image

Capturing
Macro Image
Optical conf.: <current> Objective: 4: 20x

Scanning
Optical conf.: <current> Objective: 2: 40x

Area
Large image area in XYZ Overview
5 x 4 fields, 5744 x 4302 pixels, 1,87 x 1,4 mm, 47 MB of memory

Close active shutter during stage movement
 Correct XY Offset between Macro and Scanning Objective
 Wait after Stage Movement: 0 ms

Stitching
Overlap: 10 %
Stitching via: Blending
 Image registration

Shading Correction
 Off (not available) Automatic Shading Correction

Create large image
 Store single images
 Create both

Save large image to file
 Save to Auto capture folder
Format: nd2
Filename: C:\Data\Biond\002-001\A4_large_001.nd2

Storage for single images:
Folder: C:\Data\Michal\test capacity PC memory\Large Image
Format: tif

Combine with
 None
 HDR Steps: 5
 Z Series Count: 5
 Max IP Step: 0,9 µm ← 0,900 µm
EDF
Z-drive: Ti2 ZDrive
Order: Z series per channel

Multichannel capture
Setup
+ Add [Icons]
Opt. Conf. Name Comp. ...
 Spin...:561nm ... 561nm
Stitch using channel: 561nm

Focus
 Focus manually at start
 Use Focus Surface
 Use step-by-step focus Setup...
 Focus manually
every 1 field
after 3 mm distance
 Skip frames without sample
 Manually locate sample and background at start
Optical conf.: Spinning Disk:561nm

Scan Close Help

You can image several Z plans as Z stack or max projection +

Define area boundaries

Channel setup

Define stitching method

You can keep both raw and stitched images

Nis Elements- stage overview tools

Options to define area

You can define your scan area by an ROI (1) ; four border points (2) ; number of FOVs (3)

The image displays three panels from the NIS Elements software interface, illustrating different methods to define a scan area.

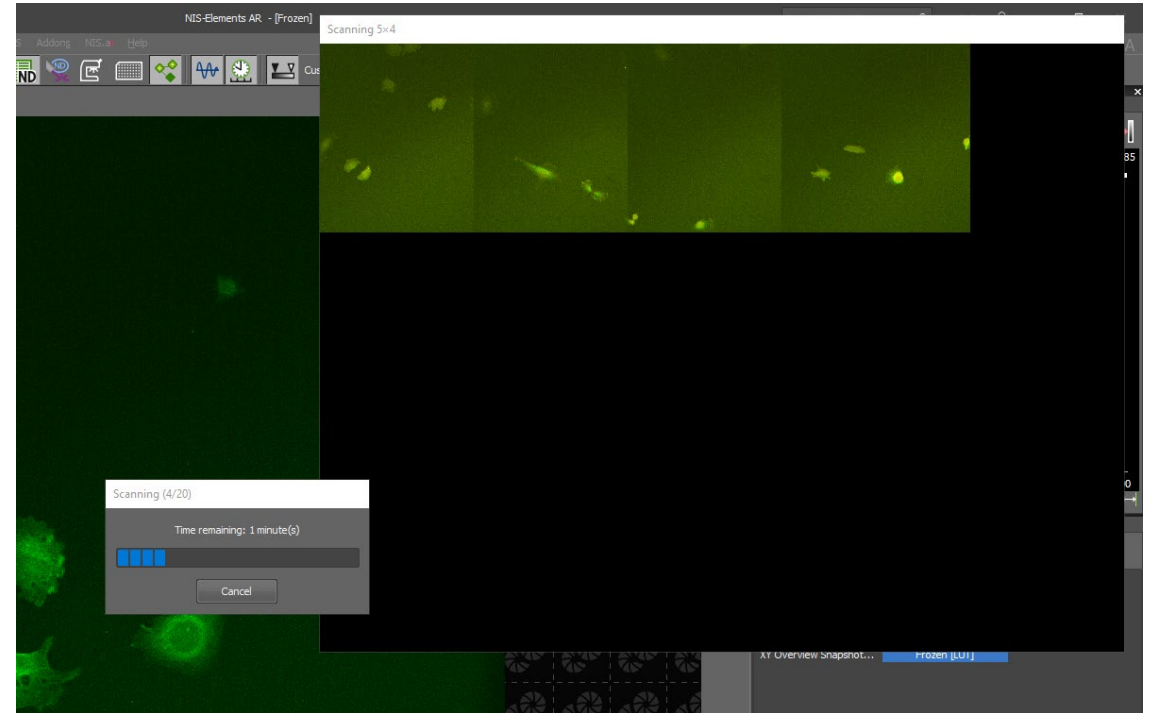
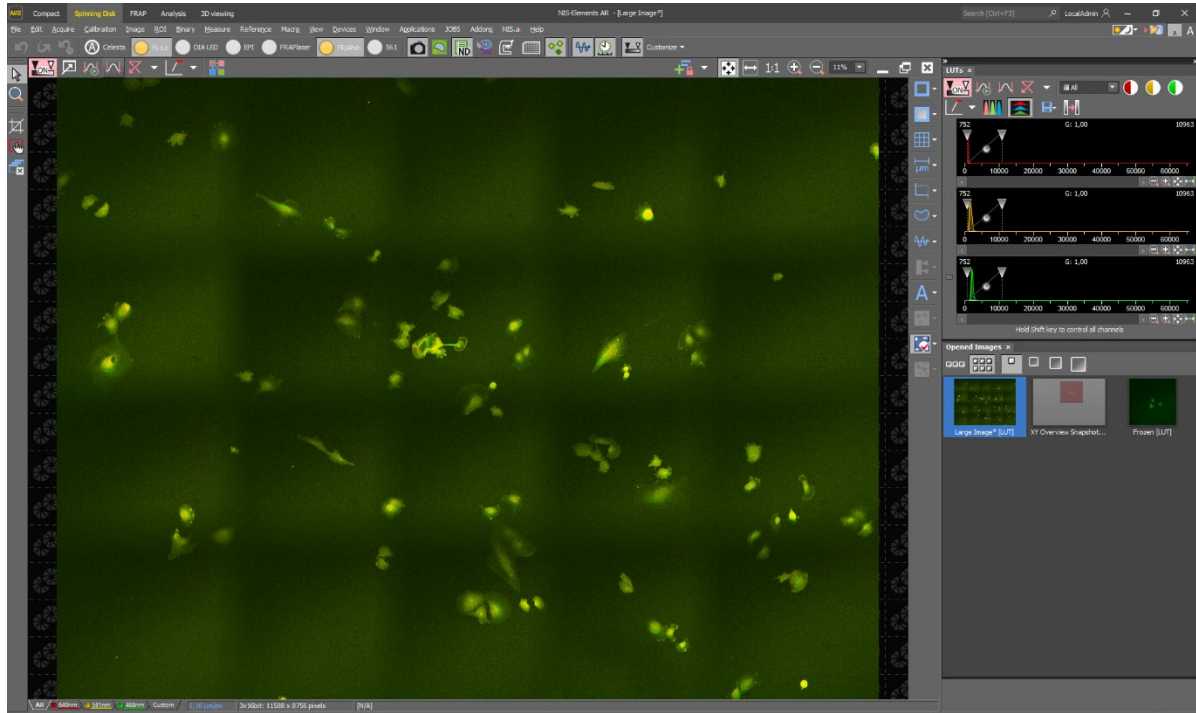
Panel 1 (Top Left): Shows the 'Area' settings for a 3x3 field scan. The 'Left, top, right and bottom limits' dropdown is highlighted with a red '2'. The scan parameters are: 3 x 3 fields, 3052 x 2949 pixels, 0,99 x 0,96 mm, 17 MB of memory. Navigation buttons for X and Y axes are visible.

Panel 2 (Bottom Left): Shows the 'Area' settings for a 4x4 field scan. The 'Number of fields in X and Y' dropdown is highlighted with a red '3'. The scan parameters are: 4 x 4 fields, 5032 x 5032 pixels, 1,64 x 1,64 mm, 48 MB of memory. 'Fields placement' options include 'Around the current position' (selected), 'Store single images', and 'Current position is at top-left corner'.

Panel 3 (Center): Shows the 'Scan Large Image' dialog box. The 'Area' dropdown is set to 'Large image area in XYZ Overview' and is highlighted with a red '1'. A red box around this dropdown is labeled 'Define area boundaries'. The dialog includes sections for 'Capturing', 'Scanning', 'Stage Overview with Macro Image', 'Combine with', 'Multichannel capture', 'Stitching', 'Shading Correction', and 'Focus'. The 'Format' is set to 'nd2' and the 'Folder' is 'C:\Data\Michal\test capacity PC memory\Large Image'. A play button is visible at the bottom left of the dialog.

Nis Elements- stage overview tools

Your scanned large image (here in 3ch) can be further used as a map for navigation- right click and “add this point to ND acq”



Nis Elements- Stage Overview tool

Create focus map

Time Measurement x

Measure Stop Step [Icons] Export

Left: Cy5 Right: none ROIs Channels

ROI: Define... Open... Save... Clear... Display: Mean Int. in ROI(s) Measurement: [Icons]

ROI #1 - Stimulation [Color] Graph Data Measurement Records: empty not running

XYZ Overview x

ND Acquisition Overview Focus Surface PFS Surface Document Overview

[Icons]

Zoom In

Create Focus surface- add position with different Z, the software will interpolate trend

Interpolation method Move Stage to Selected Point

3034,5 3028,1 #1 #2 #3 #4 #5 #6 #7 3028,1

Graph Data AOI: 57.00 mm x 37.50 mm, Scan Area: 0.07 mm x 0.07 mm

XYZ Overview x

ND Acquisition Overview Focus Surface PFS Surface Document Overview

[Icons]

You can apply the same with PFS

Zoom In

3037,7 3035 3032,5 3030 3027,6

Interpolation method Move Stage to Selected Point

3034,5 3028,1 #1 #2 #3 #4 #5 #6 #7 #8 #9 #10 3032,5

Graph Data AOI: 57.00 mm x 37.50 mm, Scan Area: 0.07 mm x 0.07 mm

Nis Elements- Triggering

The system is fully triggered: light source-emission filters-camera-piezo stage

You can define your acquisition to be triggered, meaning running very fast acquisition:

#	Line	Power	Exposure	Imaging
✓ # 1	set EM Wheel (FW)	2 (FF02-511/20)	50 ms	<input type="checkbox"/>
✓ # 2	Line: 488 nm	10 %	50 ms	<input checked="" type="checkbox"/>
✓ # 3	set EM Wheel (FW)	3 (FF01-595/31)	50 ms	<input type="checkbox"/>
✓ # 4	Line: 561 nm	12 %	50 ms	<input checked="" type="checkbox"/>

Advanced Settings

- Use Focus Offset (Piezo Z)
- Use more lines for one channel
- Don't sync power (allow same lines with different power)
- Use non-imaging channels

Use these devices:

- 561 (Shutter)
- FRAPsh (Shutter)
- DM Wheel (Filter Wheel)
- EM Wheel (Filter Wheel)
- EX Wheel (Filter Wheel)
- Follow Exposure
- Follow Exposure

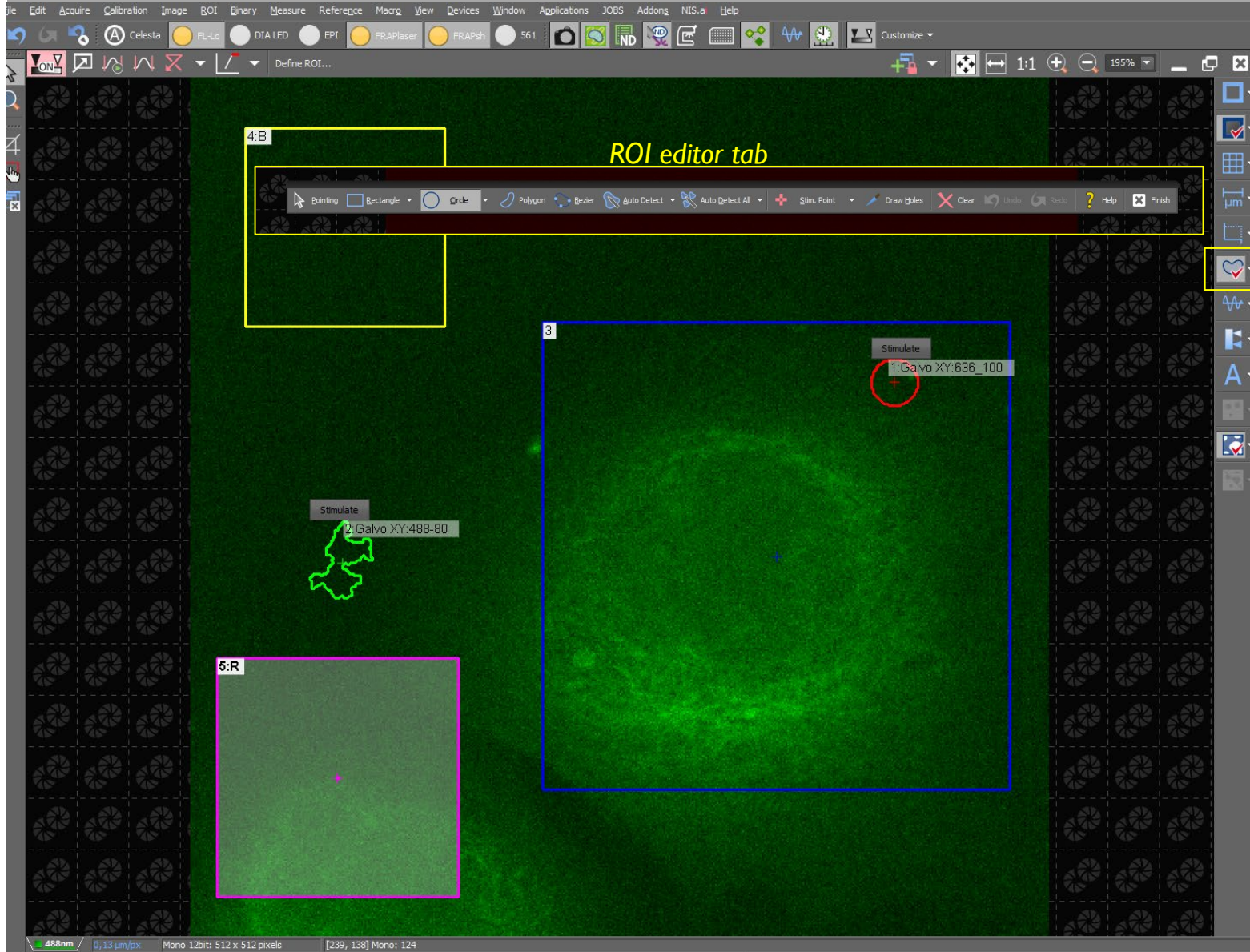
Trig is saved to your OC- don't forget to update

**You can set the exposer time and power per line
Make sure to uncheck "imaging" for the filter wheel line**

Once you enabled triggering- it will be in your channel when running ND experiment, no need for extra definitions.
If you run a Z stack- chose "triggered piezo" for fast imaging

Nis Elements- FRAP

Image view



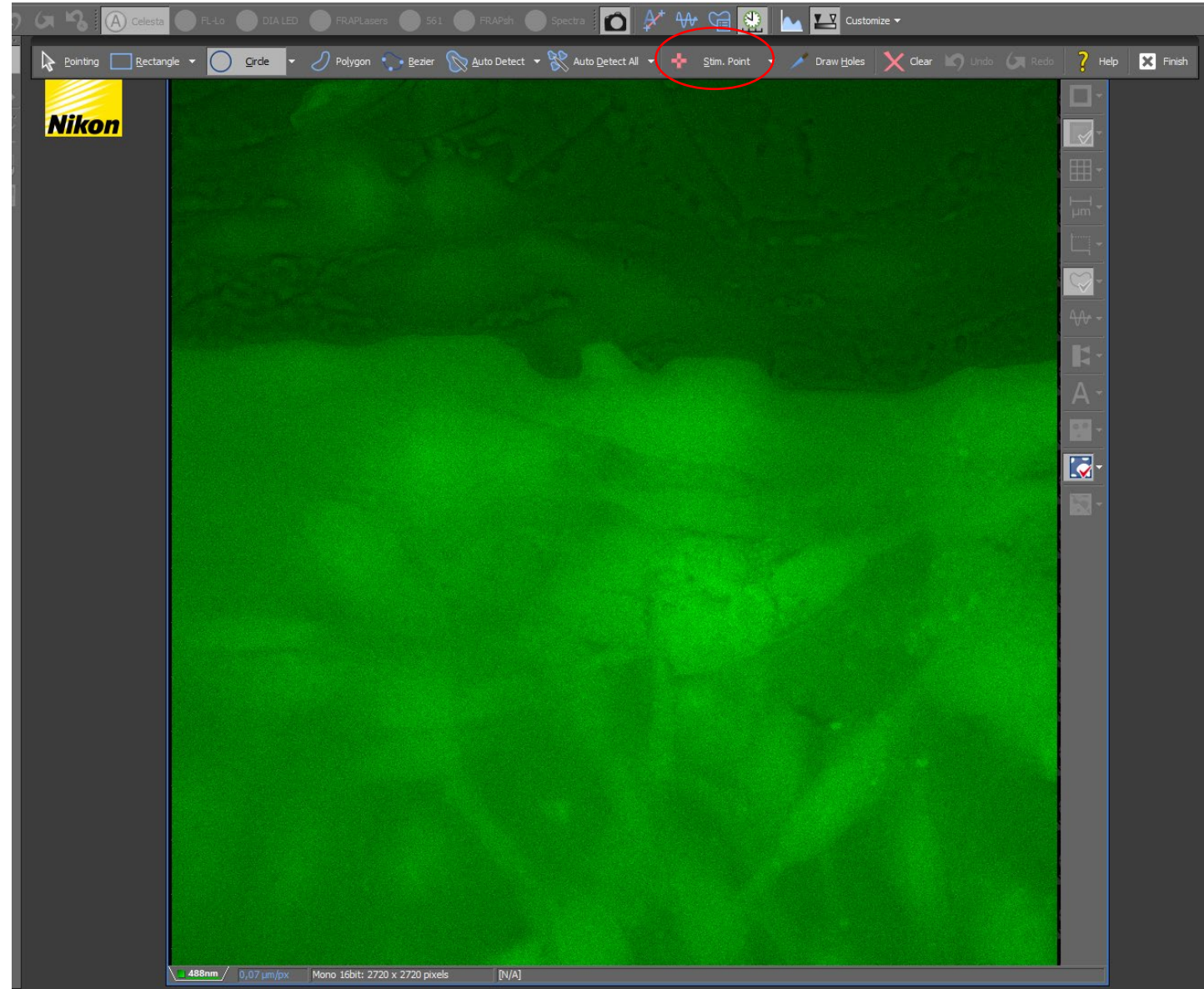
- 1, **Snap** an image
- 2, You can **set ROIs** with different sizes/contour:
A fixed shape, auto-detect, or point*

*For point stimulation you need to be on "Live" mode

- 3, **Assign each ROI :**
 - Stimulation-** assign FRAP laser and intensity pre-set
 - Background**
(no signal)
 - Reference**
(signal but not bleached)

To mark a stimulation point on your sample-

- 1, Make sure you are in live mode
- 2, right click on ROI tools and mark “ROI toolbar” to display the bar on top of your image
3. In it you can see the red cross for stim point, place it on the desired location. Clicking “stimulate” on top (without releasing the mouse) can allow you to test the time required for bleaching



Nis Elements- FRAP

Acquisition control- Simultaneous Acquisition/Bleaching

Galvo XY

Presets: 405-25, 488-25, 488-80, 561-100

1: 405 nm, 50 [%]
2: 488 nm, 80 [%]
3: 561 nm, 51,3 [%]
4: 636 nm, 52,6 [%]

Dwell Time: 200 µs
Stimulation / Scan Time: 1073,6 / 1478,8 ms

Stimulate ROI 1

Calibrated [Kinetix]

On the Fly

- 1, Enable the lasers
- 2, Save pre-sets
- 3, Optimize dwell time
- 4, Click on "Stimulate ROI"

ND Stimulation

- 1, Set the total time of acquisition
- 2, Set bleaching/Stimulation:
Wait : Delay from start of Acq
Position: Static
Duration: Dwell time as optimized
Stim.Conf: your chosen pre-set*

*Make sure it fits your ROI assignement.

ND Stimulation

Experiment: ND Stimulation

Save to File
Path: C:\Data\Michal\20230726\frap check
Filename: check_roi_488_simultaneous003.nd2 ND2

Time schedule (Kinetix)
Acquisition: Interval: No Delay, Duration: 1 min, Loops: 300

Stimulation/Bleaching: Wait: 500 msec, Position: Static, Duration: 1 sec, Stim. Conf: 488-80 (Galvo XY)

Perform Time Measurement (0 ROIs, 1 stim./bleaching ROIs)

Enable lasers for acquisition

Load Save Remove

1 time loop Run now

Time Measurement

Measure Stop Step

Left: 488nm Right: none

ROI: Define... Open... Save... Clear... Display: Mean Int. in ROI(s) Measurement:

ROI #1 - Stimulation

Intensity vs Time [s] graph showing a peak around 2.5s and a drop to zero at 4.0s.

Graph / Data / Measurement / Records: 59 (4.6 KB) not running

Nis Elements- FRAP

Acquisition control- Simultaneous Acquisition/Bleaching

You can Image while Bleaching (Except 561)

The screenshot displays the NIS Elements software interface for FRAP acquisition control. The interface is divided into several panels:

- ND Simulation:** Shows experiment settings, including the path to the simulation file and acquisition parameters like interval and duration.
- XYZ Overview:** Provides a 3D view of the microscope stage and the position of the ROI.
- Macro Panel:** Contains the 'Acquisition' section with a 'Live' button and various acquisition options.
- Time Measurement:** Displays a graph of intensity over time, showing a bleaching step and recovery.
- Graph / Data:** Shows the current acquisition parameters, including the ROI size and the current intensity.
- Colocalize:** Lists the channels being acquired, including the 561 nm channel.
- Fluorescence Image:** A large central window showing a green fluorescence image of a cell, with a red ROI overlaid.
- Opened Images:** A list of captured images, including the current frame and previous frames.

The 'Colocalize' panel shows a list of channels with their respective wavelengths and intensities. A red box highlights the '561' channel, indicating it is the channel being bleached. The 'Fluorescence Image' shows a bright green spot, indicating the presence of the fluorescent probe.

Nis Elements- FRAP

Acquisition control- Sequential imaging

You Can also set sequential ND experiment:

Acquisition
Bleaching
Acquisition

The screenshot displays the NIS Elements software interface for configuring a sequential ND experiment. The main window is titled "ND Sequence Acquisition" and shows the following details:

- Path: C:\Data\Michal\20230726\frap check
- Prefix: define_run_488_636
- Timelapse:
- Sequence Definition table:

Action	Description
#1 ND Acquisition	Time(1 sec), Lambda(2)
- Merge ND files if possible:
- Buttons: Load, Save, Remove, Run Now

The "Time Measurement" window is also visible, showing:

- Measure, Stop, Step buttons
- Left: Cy5, Right: none
- ROI: Define..., Open..., Save..., Clear..., Display: Mean Intensity
- ROI list:

ROI	Color
#1 - Stimulation	Red
#2 - Stimulation	Green
- Graph, Data, Measurement tabs

The "ND Sequence Acquisition" dialog box is open, showing:

- Experiment: [Empty]
- T: [Empty], λ: [Empty]
- Order of Experiment, Timing, Record Data... buttons
- Time Schedule table:

Phase	Interval	Duration	Loops
<input checked="" type="checkbox"/> #1	No Delay	1 sec	???
- Close Active Shutter when idle: Use PFS:
- Switch Transmitted Illuminator off when Idle (0.01 s):
- Events..., Advanced >> buttons
- Buttons: Load, Save, Remove, OK, Cancel

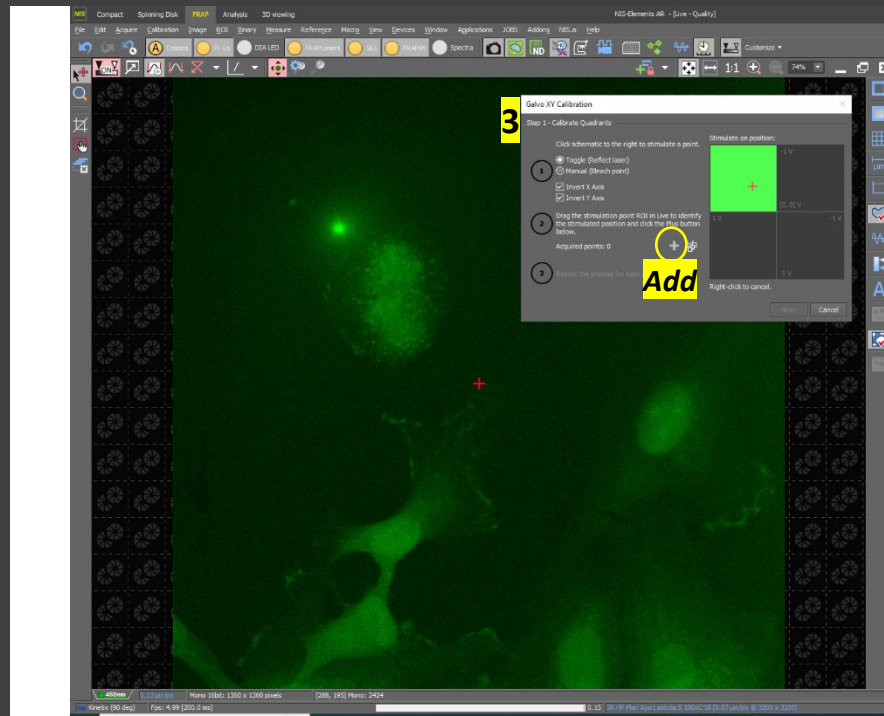
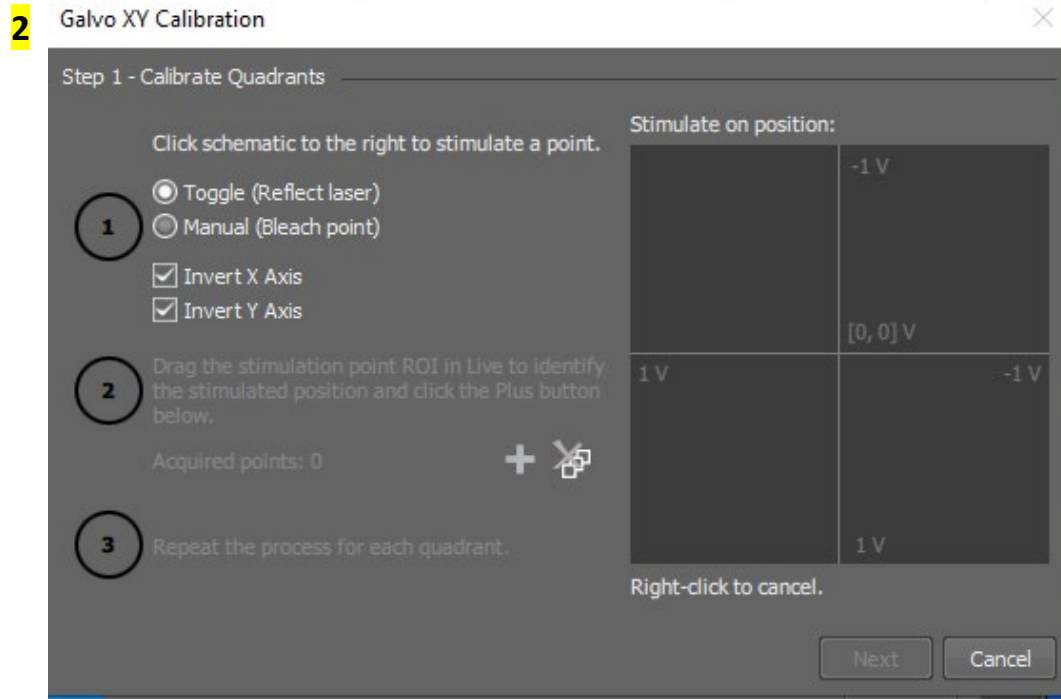
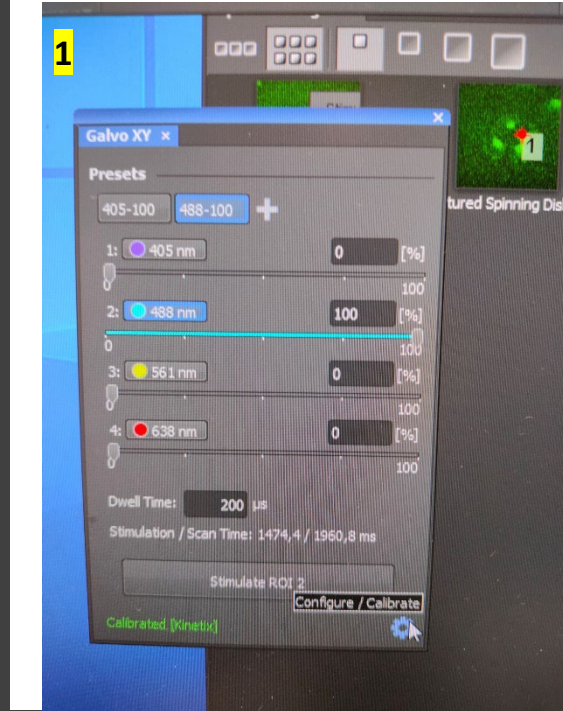
ng to Ring Buffer

ting and Photoactivating

ulator Setup

To calibrate FRAP mirrors:

- 1, go to configure/cal settings in GalvoXY panel
- 2, set on Invert
3. Click on one of the four quarters to pick a stimulation location that presents on your live view the stim point. Drag the red cross on it and then click add. Do this for the other quarters as well and repeat for accuracy



Shut down

4. PC



8. Ni Controller



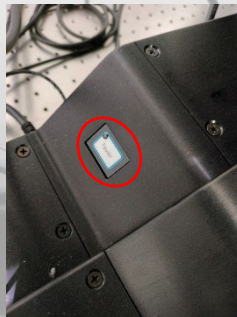
5. FRAP Ctrl



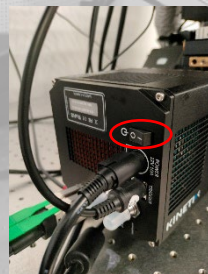
On table,
behind



6. SD unit



7. Camera



9. main
switch



1. **Save your data** locally and Copy to Bulk folder.
Note! All local data is erased monthly to keep memory free
2. ESC to minimal Z position
3. Remove your sample; Clean **Objective**; shift to 10X

Don't forget to turn off the
chamber light

