



BioFrontiers Institute
Advanced Light Microscopy Core
UNIVERSITY OF COLORADO

Innovation without boundaries

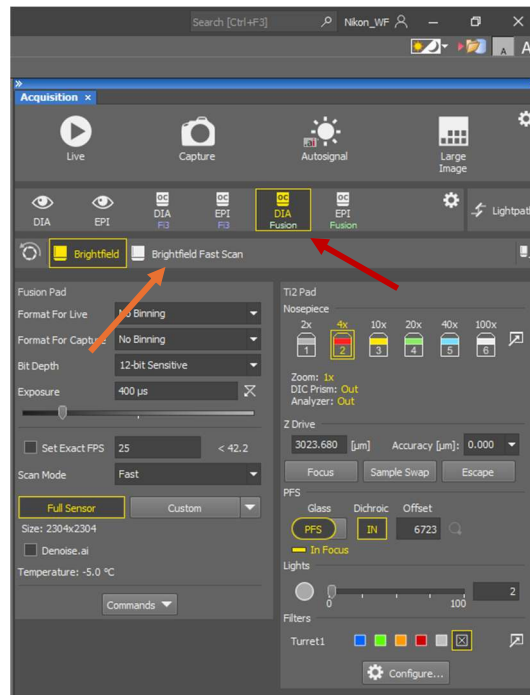
Nikon Sample Navigation

How to use Nikon Sample Navigation
to quickly scan your sample and define ROIs for imaging

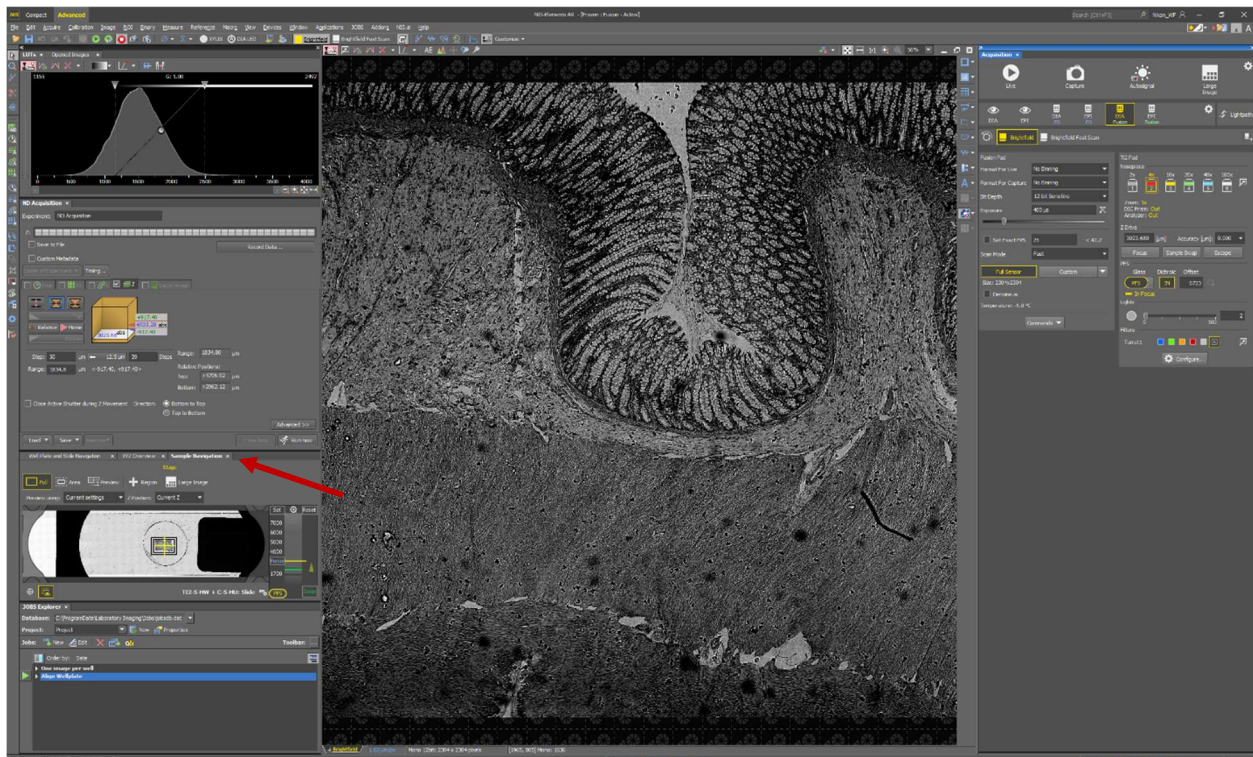
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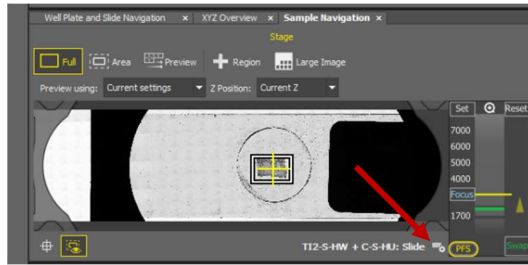
advancedimaging.colorado.edu

1. Turn the microscope on and open NIS Elements.
2. Put your sample onto the stage and bring your sample into focus (with PFS if possible) with the 4X objective.
3. Select the *DIA – Fusion: Brightfield Fast Scan* optical configuration (quickest way to use *Sample Navigation*). Make sure the exposure time is very short ($\sim 400 \mu\text{s}$ or less) to scan quickly.

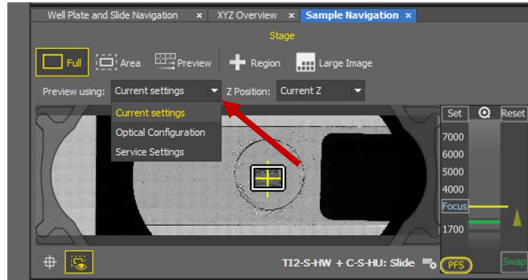


4. In the *Sample Navigation* tab, change the sample holder, if necessary. Most people use the *T12-S-HU, Universal Holder: Slide* when imaging slides.

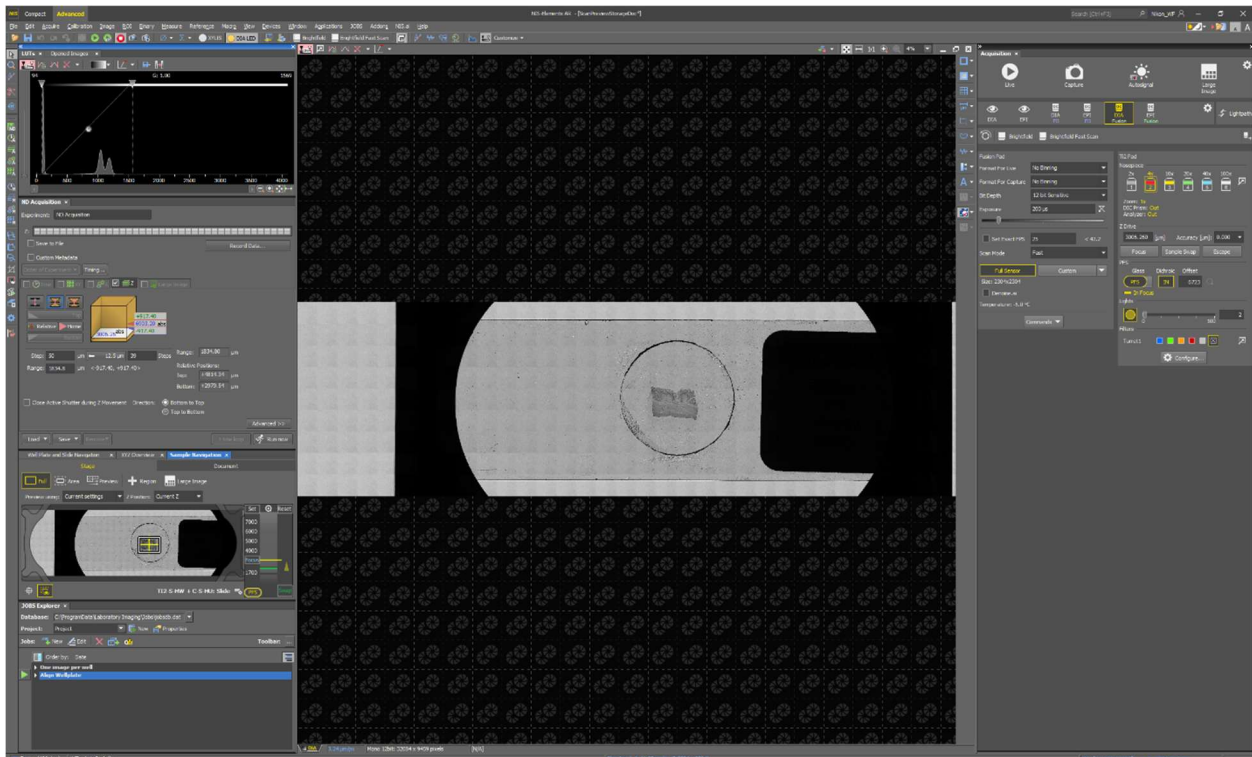
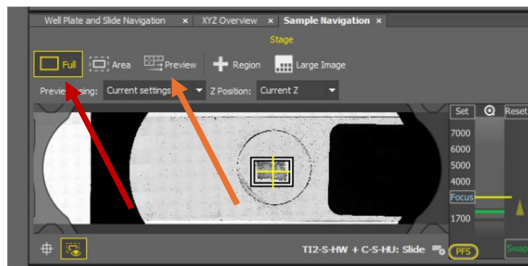




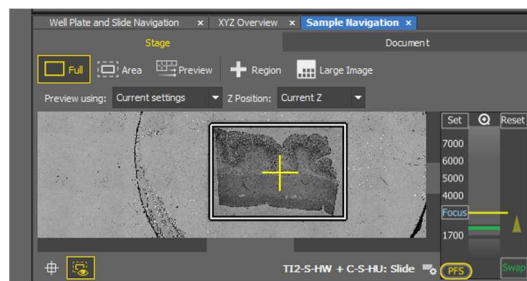
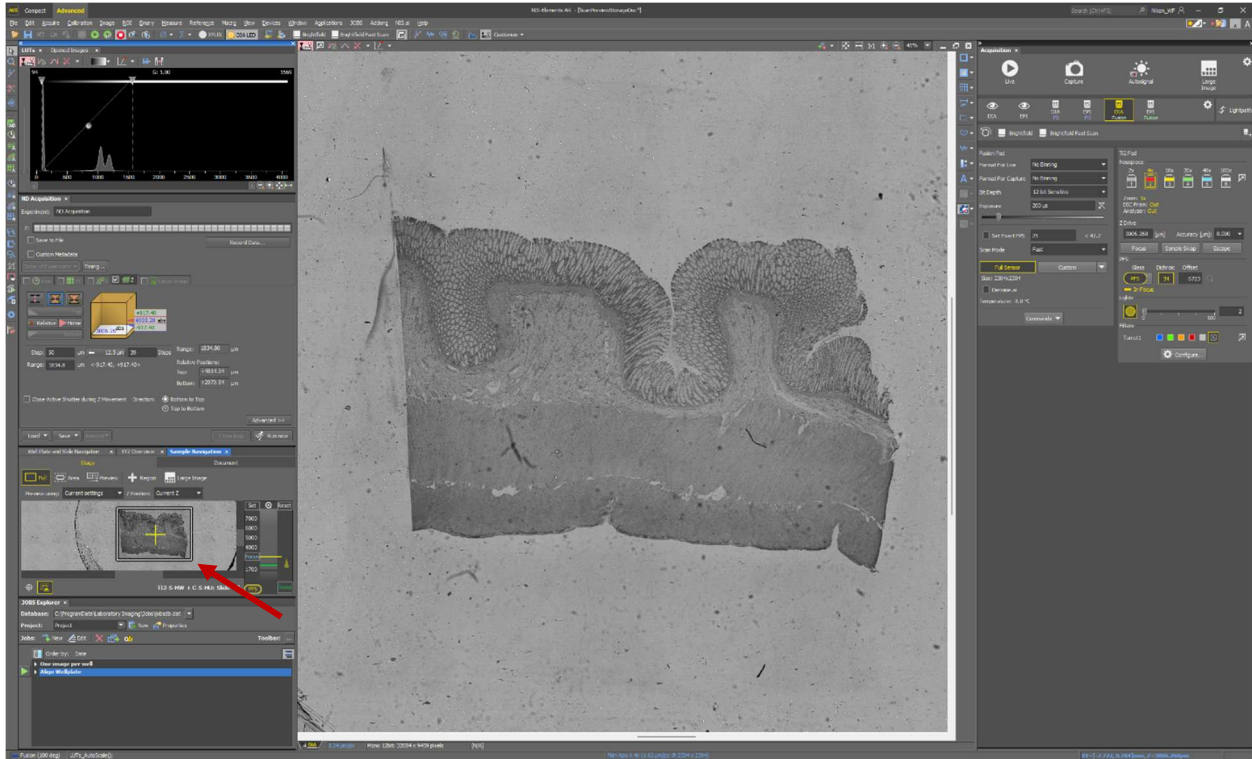
5. Then, in the *Preview using:* drop-down menu, select *Current setting*.



6. In the *Sample Navigation* tab, click the *Full* icon and the *Preview* icon to scan your full sample quickly at 4X in brightfield.

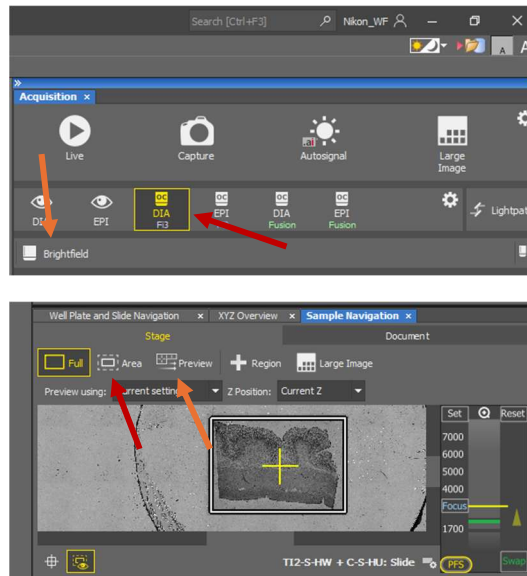


7. You can zoom into this scan on the main screen or in the *Sample Navigation* tab (using the mouse wheel) to see the sample at higher resolution.
 - a. In the *Sample Navigation* tab, click the white rectangle to adjust your scan area.
 - b. Click and drag the now red rectangle to move it around.
 - c. Click on the white vertices to shrink or expand the scan area.
 - d. Right-click the rectangle to confirm.

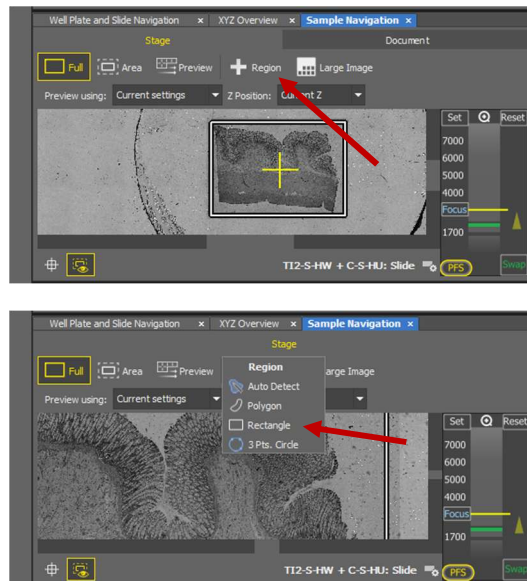


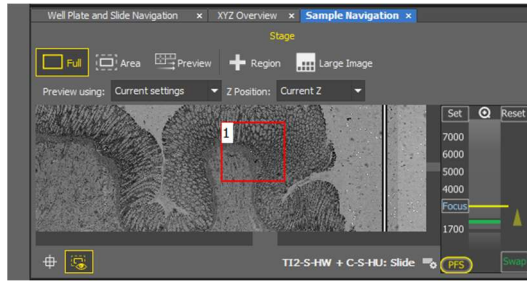
8. In the *Sample Navigation* tab, once you define the large scan area, you can scan again in a different optical configuration. Try to keep the exposure times low (~ 5 ms or less) for a quicker scan.
 - a. In the *Acquisition* tab, select your new optical configuration.

- b. In the *Sample Navigation* tab, ensure that *Current settings* is selected in the *Preview using:* drop-down menu.
- c. Click the *Area* icon to scan just the defined scan area and not the full area.
- d. Click the *Preview* icon to begin the scan.

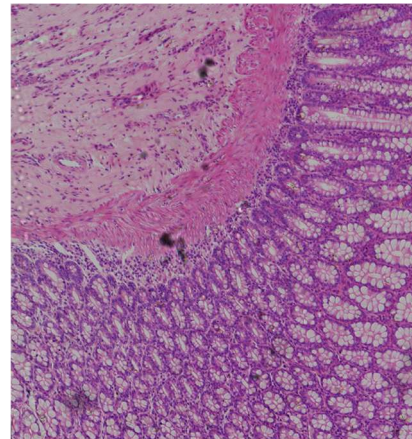
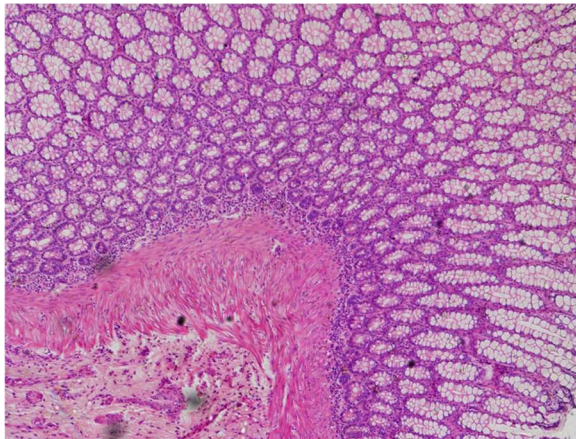
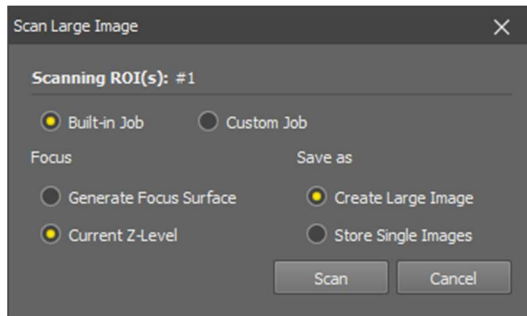
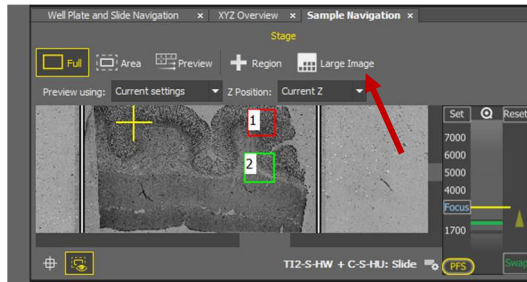


9. You can zoom into this scan on the main screen or in the *Sample Navigation* tab (using the mouse wheel) to see the sample at higher resolution.
 - a. In the *Sample Navigation* tab, click the *Region* icon to add a Large Image region.
 - b. Select the region of interest type.
 - c. Draw your region of interest. Right-click to confirm.





10. Repeat Step 9 and once all your regions are drawn, in the *Sample Navigation* tab, click the *Large Image* icon and click *Scan*.



11. Make sure to save your images!