



Cryo-EM Merit Badge Independence Checklist

A Collaboration Among the NIH-Funded Cryo-EM National Centers

Screening with Leginon Independence Test Evaluator Checklist – CryoEM Merit Badges

Trainee: _____

Start Leginon software

- ☐ ☐ Verify NCCAT Leginon client is open on microscope computer. If not, start client.
- ☐ ☐ Start Leginon session on Leginon computer.
 - ☐ ☐ Correct holder selected.
 - ☐ ☐ Proper session naming: microscope - proposal ID - misc (optional).
 - ☐ ☐ Correct project selected:
 - ☐ ☐ Proper clients selected:
 - ☐ ☐ Correct C2 size set:
- ☐ ☐ Start appropriate Leginon app.
- ☐ ☐ Load Presets from most recent session, and verify they look reasonable.

Load grid and find eucentric height

- ☐ ☐ Load grid using TEM user interface.
 - ☐ ☐ Check that column valves are closed.
 - ☐ ☐ Check that objective is out.
 - ☐ ☐ Check that screen is inserted.
- ☐ ☐ Open column valves after new grid is loaded onto the stage.
- ☐ ☐ Find a grid square to do eucentric height.
 - ☐ ☐ Grid preset sent to microscope.
 - ☐ ☐ Screen inserted.
 - ☐ ☐ Move to visible square with support film using hand panel stage movement joystick or TUI navigation.

Imaging

- ☐ ☐ Eucentric height is found using the Z_Focus node in Leginon
- ☐ ☐ Atlas acquired: opened settings to give sensible label and radius, click calculate, then acquire.
- ☐ ☐ Square targets selected in the square targeting node and submitted.
- ☐ ☐ In Hole Targeting node: Settings are checked to queue at this step (if desired), allow for user verification of targets, and automated hold finder is skipped (if desired).
- ☐ ☐ Acquisition and focus targets set and submitted in Hole targeting node.
- ☐ ☐ Verify that user can “unselect” targets.
- ☐ ☐ Queue is submitted **AFTER** all hole image targets are selected (if queueing at this step).
- ☐ ☐ Exposure_Targeting node settings are checked and set to “Allow user verification”, “skip automated hole finder” and uncheck “queue up targets” (if desired)
- ☐ ☐ Acquisition and focus targets set and submitted in Exposure targeting node (optional: if queueing, queue is submitted **AFTER** all exposure image targets are set.
- ☐ ☐ Once all selected square targets are finished, select additional targets in the Square Targeting node and repeat, or load new grid.
- ☐ ☐ Verify user can pause a node and abort a queue.
- ☐ ☐ Repeat for all grids.



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Changing grids

- ☐ ☐ Verify Legion is not actively imaging.
- ☐ ☐ Close column valves.
- ☐ ☐ Insert screen.
- ☐ ☐ Ensure objective aperture is out.
- ☐ ☐ On TEM User Interface "Autoloader" panel, click on grid and then click "load" button.
- ☐ ☐ Wait for autoloader to finish changing grids before proceeding.

Ending session

- ☐ ☐ Alert staff when finished screening for proper shut down.

Supervising Staff 1: _____

Date: _____

Supervising Staff 2: _____

Date: _____