

Screening with Legikon Independence Test Evaluator Checklist – CryoEM Merit Badges

Supervising Staff: _____

Date: _____

Trainee: _____

Start Legikon software

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

Load grid and find eucentric height

- Load grid using TEM user interface.
 - Checked that column valves are closed
 - Checked that objective is out
 - Checked 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

Evaluator Notes:

Imaging

- Eucentric height is found using the Z_Focus node in Legikon
- Atlas acquired: opened settings to give sensible label and radius, clicked 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 queuing, 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 that user can pause a node and abort a queue.
- Repeat for all grids.

Evaluator Notes:

Changing grids

- Verify Legimon 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.

Evaluator Notes:

Ending session

- Alert staff when they are finished screening for proper shut down.

Overall Evaluator Notes:
