

Microscope Usage HITACHI HT-7700 Instrument Location (Room E116 – Cherry Logan Emerson Hall)

Important

- This protocol is intended as instruction guidelines for users. It assumes previous user training and approval by **Robert P. Apkarian Integrated EM Core (IEMC)** staff.
- This protocol requires knowledge of the controls at the microscope.
- Microscope alignments will be carried out by **IEMC** staff unless the user has been properly trained and approved to do it.
- Please contact IEMC staff for assistance in case of any troubles.

Checklist before you start working on the Microscope:





- High voltage is on: HV on light in the HV/Filament operation area of the Hitachi TEM system control window is green.
- High voltage is usually 80kv. If another voltage is needed contact IEMC personnel for assistance.



• There is liquid nitrogen in the cold finger

• Make a log book entry with the time indicated on the black hour meter







Load Grid onto Grid holder:



Load grid holder into the microscope:



- Ensure that the stage position is at X=0, and Y=0 and that the stage is not tilted (alpha=0 or close to 0)
- If necessary, activate the Reset button by checking the box next to it. Then, click on it to take all coordinates back to 0
- Failure to do this may cause damage to microscope components
- Stage operation 23 Holder Speed Function Offset Present HT7700-SS 0 μm 🗹 -40 0 μm 🗹 280 <u>.</u> .3 deg 🗹 0.0 α Start α 🔺 Stop No. α Memory 2 Delete 5 Delete all ○ Slow ○ Mid ● Fast Reset
- Set the grid holder in the specimen exchange chamber, carefully aligning the position of the pin.
- Make sure that the grid holder is fully inserted until you see the red light turn on

• Turn the specimen chamber evacuation switch to EVAC







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• Once the vacuum level reaches the appropriate level for insertion (Evacuation completed), the green light will switch ON and a loud beeping noise will sound.

• Once the green light is on turn the grid holder clockwise 30 degrees. The holder inserts itself up to the retraction point due to the vacuum. Do not push the holder towards the microscope

• Turn the grid holder counterclockwise 15 degrees. The holder inserts itself up to the specimen viewing position due to the vacuum. Do not push the holder towards the microscope.



Imaging your sample:



• If the HV is not ON. Click the HV on button in the HV/Filament operation area of the Hitachi TEM system control window.	Hitachi TEM system control File Operation Function Window About File Operation Function Window About File Operation Function Window About
 Alternatively, the same HV on button is available on the HV/Filament operation window Click on Filam. on button in the HV/Filament Operation window to turn on the filament. Click on Beam on in the HV/Filament Operation window to turn on the beam. 	HV/Filament operation Image: Second constraints HV Filament Acc. voltage preset(kV) 80.0 Acc. voltage present(kV) 80.0 Filament voltage preset(V) 16.8 Filament voltage preset(V) 16.8 Filament voltage preset(V) 0.0 Muto Manual Emission current preset(µA) 10.0 @ Auto Manual Bias voltage preset(V) 965 Beam on Bias voltage present(V)
• Press the Lens Reset button on the control panel	



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Adjust Z height:

- Set the magnification to 20Kx, using the Magnification knob on the control panel
- Press the WOB (wobbler) button on the control box





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• Use the screw under the specimen holder to adjust the Z height until the image stops wobbling



Objective Aperture Stigmatism Correction:

Find a hole or other round object on your sample using the Stage Control Trackball on the left control panel OPERATION PANEL Increase the magnification to 80 . . . KX or higher, using the Magnification knob on the control panel -----Find a hole or other round object on your sample Slightly over focus the object using the Focus knob on the control panel to obtain a fringe around the hole Press the OS (Objective Stigmator) • button on the control panel OPERATION PANEL Use the X and Y knobs to correct . . . stigmatism until the fringe is even all around the hole Press BH button on the control panel to get out of the OS mode. Focus on a hole to ensure that the image is sharp. Alternatively, using the FFT mode • File View of the CCD camera (Multi viewer operation window), astigmatism can be corrected by making the center of the



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FFT as round as possible. Activate the OS mode and adjust the stigmatism X and Y while changing the focus on the image

Camera and image capture operation.

 Click on the Run button in the CCD Operation window to start the camera redout Adjust the histogram to center it in the window using the Brightness control knob in the control panel 	CCD operation File View Image Maintenance Image Auto LUT Black Image Black Image Image </th
 Center your area of interest using the Stage Control Trackball on the left side of the control panel Adjust focus using the Focus knob on the control panel 	
 Click freeze to capture your image, if you like it, click save Click run and search for another area or stop to return to the normal screen TEM image Always return to this mode when finished or lowering magnification to search another area of the grid 	CCD operation File View Image Maintenance Image Auto LUT Black White Gamma Image Image </td

When you are done.



 Click on the Filam. off button in the HV/Filament Operation window to turn off the filament. Click on Beam off in the HV/Filament Operation window to turn off the beam. Do not turn off the HT 	HV/Filament operation ⊠ HV Filament Acc. voltage preset(kV) 80.0 Acc. voltage present(kV) 80.0 Acc. voltage present(kV) 80.0 Filament voltage preset(V) 16.8 Filament voltage preset(V) 16.8 Filament voltage preset(V) 0.0 Auto Manual Emission current preset(µA) 10.0 Bias voltage preset(V) 965 Beam on Beam off
 Ensure that the stage position is at X=0, and Y=0 and that the stage is not tilted (alpha=0 or close to 0) If necessary, activate the Reset button by checking the box next to it. Then, click on it to return stage to the center position Failure to do this may cause damage to microscope components Remove the holder from the microscope by inverting the instructions 	Stage operation Image: Stage operation Holder Speed Function Offset Pretent HT7700-SS x 0 µm 280 0 x 0 µm 280 0
 indicated for holder insertion Remove your grid from the holder and reinsert the holder into the 	



microscope

hour meter

- Make log book entry with the current finish time indicated on black