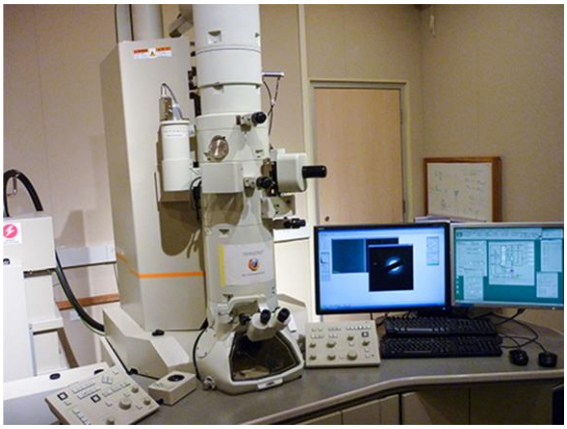
Center for Nanoscale Systems
Harvard University
FAS / SEAS

Standard Operating Procedure

HAR 027 JEOL 2100

Edited (02/20/2020)



Only users who have completed training by CNS staff are authorized to use this tool.

Emergency

In the event of an emergency, contact the nearest CNS staff member.
In *urgent* cases, such as

- fire or medical: call 911
- public safety: call Campus Police 5-1212
- all other: call University Operations center 5-5560

Safety

Instrument specific safety information:

1. The JEOL 2100 is equipped with high voltage shielding, which must not be tampered with.
2. In case of a dire emergency (eg sparks, fire) leave the room immediately, put a sign on the door and seek assistance from staff and/or the University Operations center. Do not attempt to shut down the instrument.
3. Follow liquid nitrogen training protocols and wear the appropriate personal protective equipment when filling the anti-contamination device (ACD).

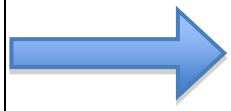
Contact

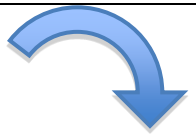
Please notify a staff member immediately if you encounter problems on the instrument. For assistance, please contact

1. Adam Graham Primary Agraham@cns.fas.harvard.edu
2. Jules Gardener Alternate Jgardener@cns.fas.harvard.edu

Acknowledge

CNS should be acknowledged in any publication resulting from work done using CNS facilities, staff or other resources: *This work was performed in part at the Center for Nanoscale Systems (CNS), a member of the National Nanotechnology Coordinated Infrastructure Network (NNCI), which is supported by the National Science Foundation under NSF award no. 1541959. CNS is part of Harvard University.*

Sample rod removal						
Set switch to **AIR**	Smoothly pull rod OUT 	Rotate rod counter-clockwise to the stop. 	Pull out slightly 	Rotate to the *second* stop  ***Wait*** for sound of valves	STOP ***Wait*** for the Specimen /PiG4 status to read 238 µA or higher	It is now safe to completely remove the sample rod. 

Sample rod insertion							
With Switch Set to AIR	Align *pin* on rod to *slot* on goniometer	Push rod IN  ***Wait*** for sound of valves; set switch to **PUMP;** Amber light appears	STOP Let go of sample rod & ***wait*** for Green light (~34µA)	Rotate to the second stop 	Guide in slightly. 	 Rotate rod clockwise to the stop	*Slowly* guide completely into column 

How to Use the JEOL 2100

Basic TEM-mode Alignment at 200kV instructions

Startup

1. Log into Clean System and record the filament hours (counter on big tower).
2. Look at vacuum gauge and make sure it's to the left of the pencil line at 2.5. Ensure that the scale selector is set to 10^{-5}Pa.
3. Make sure the green light of the ion pump is on.
4. On the right monitor, make sure the pressure gauges indicate Evac Ready.
5. Turn on HT high voltage (only the first user of the day should have to do this):
   - Look at the High Voltage Control setting. Make sure the HT is off and set at 180 kV (if not, use the up/down arrows to adjust to 180kV).
   - Click HT on (you will hear a click).
   - Wait for the popup window to go away.
   - In the Auto HT menu (halfway down the window) set the target to 200 kV, step 0.05 V, time 3 sec and press START. This should take 20 minutes.
6. Put liquid N2 into the anti-contamination device (ACD):
   - Pull the heater out of the ACD.
   - Put the plastic cover over the viewing window.
   - Use a funnel.
   - Fill with LN2 until it overflows. If the ACD trap was at room temperature, then the LN2 will boil off and will need to be topped up.
   - Place the stopper in the trap opening (this keeps moisture out).

Remove the sample rod

1. Check that the filament is off.
2. Check that the correct holder is selected from the drop-down menu (eg single tilt beryllium).
3. Double click Stage Neutralize. Check that the goniometer position is at zero.
4. Set the toggle switch to air.
5. Follow the sample rod removal instructions on the sheet provided (pull back straight, twist counterclockwise, pull back slightly, twist slightly counterclockwise, **WAIT for vent**, pull out completely).
6. Don't touch the sample rod beyond the o-rings with your bare hands. Don't drop it!

Mount your sample

1. Wear nitrile gloves when mounting your samples.
2. Place the sample rod on its stand.
3. Insert the small tool and open the jaw of the rod. Remove the sample blade with tweezers and place it on the plastic holder (align the small pin with the hole on the sample holder).

4. Loosen both screws and slide the t-shaped clip to the side. There is no need to fully remove the screws.
5. Insert your TEM sample carbon side up (you can manipulate TEM grids with tweezers or the vacuum pen).
6. Slide the t-shaped clip back in place so that it clamps the grid down. Tighten both screws but do not overtighten.
7. Check that your TEM sample is held securely in the sample blade.
8. Use the tool to open the rod's jaw and replace the sample blade clip side up. Check that the sample blade is seated securely in the rod.

Insert the sample rod

1. Ensure that the toggle switch is at air. Align the pin on the sample rod with the notch in the goniometer.
2. Push the rod in until it stops (the pin goes in ~1cm beyond outside of goniometer). Wait for the valve sounds, then switch the toggle to pump (orange light comes on). If you don't hear valve sounds, then the rod's position needs adjusting because the microswitch hasn't activated - push the rod in gently and rotate slightly counter-clockwise.
3. Wait for the green light (5-10min, 34-36uA). If you don't wait you will crash the vacuum!
4. Finish inserting the rod: twist the rod clockwise, then guide in a short distance, twist again (through a larger angle) and guide in smoothly until the handle is close to the goniometer opening and valve noises are heard.
5. Check that the vacuum is below the pencil line again (gauge on big tower).

Turn on the filament

1. Make sure the filament set point is 40%. Use the arrows next to the filament value to adjust this set point. DO NOT adjust the filament max limits.
2. Make a mental note of the filament current at this time. This is the dark current.
3. Click the Filament On button. You will hear a click.
4. Once the dialog box disappears, you can slowly increase to current by clicking the up arrow once every second. Watch the beam current for spikes (reduce the filament if the beam current spikes). Currently the saturation is from 58%-61% This will change over time
5. The final current should be 4-8uA higher than the dark current (this is the bright current). If Current is higher than 10uA please get a staff member to help.

Find the beam and sample region of interest

1. Adjust the room lighting for imaging.
2. Remove the plastic viewing window.
3. Look for the electron beam on the screen. If this can be seen, then move on to the next sections.
4. If you cannot see the beam: Adjust the brightness dial and/or reduce the magnification. Translate the sample in the x-y directions using the trackball until a region of the sample you wish to image is in the center of the screen.

Set the z-height of the specimen to the eucentric height

1. Press the standard focus button (this resets the objective focus to its nominal value).
2. Adjust the magnification until features can be seen on your sample (eg 40kx). Use the trackball to adjust the x-y position if needed.
3. Use the brightness dial to adjust the illumination on the screen. The screen should be bright enough that features can be seen.
4. Press Image Wobble X. If the image moves on the screen, then the height needs adjusting.
5. Select the coarseness of z-motion (green arrows next to the z position reading). Press and hold the z up or down arrows until it the image motion stops. The motion will stop, and contrast will be reduced at the eucentric height.
6. Increase the magnification and re-adjust the z-position to stop the wobble.
7. Deselect the Image Wobble X button.

Insert and Align the Condenser Aperture

1. Set the magnification to a moderate value (eg 40-100kx).
2. Set Spot Size = 5 and Alpha = 3 using the knobs.
3. Use the brightness knob to make the beam small. Ensure that none of the selector buttons are illuminated and use the shift x and shift y dials (i.e. condenser shift) to center the beam.
4. If an aperture isn't already inserted, insert the largest condenser aperture by turning the black dial so that the biggest white dot is aligned with the black mark on the instrument (instead of the red dot).
5. Use the brightness knob to make the beam almost as big as the screen and adjust the position of the condenser aperture using its front and side knobs until the beam is centered on the screen.
6. Set the spot size to Spot Size 1.

Align the Gun

1. Set to a high magnification (eg 100-400kx). Make the beam as small as possible using the brightness dial.
2. Lower the filament current gradually until you see a dark pattern on the screen. This occurs at a filament saturation of 49-57% (these numbers change over time). Use the brightness dial to focus the pattern.
3. Press F4 to enable gun deflector mode. The shift knobs become gun shift knobs and the lower knobs (def/stig) become gun tilt knobs.
4. Use the shift x/y knobs to gun shift the beam to the middle.
5. Use the def/stig knobs to adjust gun tilt so that the pattern is symmetric and at its brightest.
6. Adjust the gun shift to get the spot in the middle again.
7. Press F4 to deselect the gun controls.

Adjust the Condenser Astigmatism

1. Push the Cond Stig button to make the def/stig dials into condenser stigmation controls.
2. Adjust the condenser stigmation until the pattern is as sharp as possible (i.e. not streaky or distorted).
3. Push the Cond Stig button again to deactivate it.

4. Increase the filament to its saturation value again using the up arrows (these can be clicked a few times per second this time).

Fine-tune the gun shift

1. Set the magnification to a moderate value, eg 50-100kx.
2. Set the spot size to 5, make the beam small using the brightness dial (it's OK if it doesn't look circular!), center the beam using the shift dials and re-center the condenser aperture (as described earlier).
3. Change the spot size to 1, make the beam small, press F4 to activate the gun controls and center the beam using the shift dials (i.e. gun shift).
4. Press F4 to deactivate the gun controls.
5. Cycle through steps 1-3 until the beam center stays stationary between spot sizes 5 and 1. Leave in spot size 1.

Adjust High Voltage Centering

1. Set the magnification to 50kX-200kX. Spread the beam so that it extends beyond the screen (using the brightness dial). Position a small well-defined feature in the center of the screen using the trackball.
2. Check the alignment of the condenser aperture (bring the beam to its smallest, center using the shift knobs, spread the beam and adjust the condenser aperture knobs if needed).
3. Some people find it helpful to bring up the mini screen at this point.
4. Press the HT Wobble button.
5. Press the Bright Tilt button.
6. Use the def/stig knobs to adjust the bright tilt until the feature stops swaying and goes in and out of focus but remains centered.
7. Press Bright Tilt and HT Wobble to deactivate these buttons. Remove the mini screen (if used).
8. Re-check the alignment of the condenser aperture and adjust if needed.

Fine-tune the alignment

1. Each alignment step affects the others. Cycle back through any of the previous alignment steps to further improve the alignment. This is particularly important if you made large changes at one or more steps.

Display an image on the PC

1. Open the Gatan digital micrograph software if it's not already open. The camera is Peltier cooled.
2. Check Camera Inserted to insert the camera.
3. Make sure Auto Survey is checked (it adjusts the histogram).
4. Press Start View.
5. Open up the Live FFT (Process > Live > Live FFT).
6. Set the brightness so that the screen is fully illuminated.
7. Flip screen up by pressing F1.
8. Adjust the Objective Focus by turning the coarse or fine knobs.

9. If the defocus is high (>300), redo the focus: flip the screen back down (F1), press Standard Focus, Image-X Wobble and the up/down (z) buttons (as described earlier).

Correct for Objective Astigmatism

1. Adjust the focus until rings can be seen in the FFT (you may need to adjust the brightness/contrast of the FFT via dragging over the histogram).
2. Press OBJ Stig.
3. Use the lower def/stig dials to adjust the objective stigmation until the FFT rings are round. There should then be no streaking in the live image as you move out of focus.
4. Press OBJ Stig to deactivate this button.
5. Use the objective focus course/fine dials to re-focus the image.

To increase the magnification of an image

1. Flip the screen down (F1).
2. Increase the magnification.
3. Adjust x-y shift and condenser aperture position (if necessary).
4. Adjust the brightness so that the beam fills the screen.
5. Raise the screen (F1).

To save images

1. Set the exposure time to “auto” or “user” (select the duration for the latter).
2. Click Start Acquire.
3. Save the image as a .dm3 file on our network drive in User Images -> JEOL 2100 -> Your folder. This can be viewed later in ImageJ or other software.
4. Do not use a flash drive.

To change between samples

1. In the Digital Micrograph software: uncheck “camera inserted” and click “stop view”.
2. Lower the screen (F1).
3. Turn filament down to 40% using the up/down arrows.
4. Turn the filament off.
5. Neutralize the stage position by double clicking Stage Neutral. Click OK on the dialogue box when complete.
6. Remove the sample rod, change the sample and re-insert the sample rod (follow the instructions in earlier in this document).
7. Turn the filament back on, initially at 40% and then increase to saturation using the up arrows (as before).
8. Bring the sample to the eucentric height (see earlier instructions).
9. Check the condenser aperture alignment and tune up if needed.
10. The system should be ready to image.

At the end of your session (if there will be more users that day)

1. In the Digital Micrograph software: uncheck “camera inserted” and click “stop view”.

2. Lower the screen (F1).
3. Set mag to 500k, Spot Size 1, Alpha 3, 200 kV (ramp up if you need to).
4. Turn filament down to 40% using the up/down arrows.
5. Turn the filament off.
6. Neutralize the stage position by double clicking Stage Neutral. Click OK on the dialogue box when complete.
7. Put plastic cover over the viewing window.
8. Remove the sample rod and your sample and re-insert the sample rod (follow the instructions in earlier in this document).
9. Leave the HT at 200kV and the filament off.
10. Log out of the CLEAN system.
11. Report any problems with the instrument to a member of CNS staff.

<u>At the end of your session (if there will not be any more users that day)</u>

1. Follow steps 1-8 above.
2. Turn HT down to 180 kV in 5kV steps over the course of a few minutes.
3. Turn the HT off
4. Make sure the sample rod is inserted.
5. Remove all apertures by turning the dials to the red dots.
6. Remove the LN2 cap and slowly insert the heating element, ensuring that the pins connect.
7. Click ON on the ACD Heat control panel.
8. Log off of the CLEAN system and add a note that you performed an ACD heat.
9. Report any problems with the instrument to a member of CNS staff.