

SEM Training Notebook

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Before you begin...

- Complete the required safety training modules on UC Learning
 - Laboratory Safety Orientation (Fundamentals) 2013
 - Hazardous Waste Management
 - Compressed Gas Safety
 - X-Ray Safety
- Submit a copy of your Training Transcript to Lab Manager
- Review the MSE SEM Policies and Regulations
- Fill out the SEM FAU Authorization Form with PI signature
- Fill out the MSE 150, 250, 309 Authorization Form with PI signature
- Receive a user name and temporary password for Faces scheduling
- Arrange a time for SEM training with Lab Manager
- Schedule a 2 hour block on Faces for your training
- Familiarize yourself with the graphical user interface (GUI) :A – D
- Familiarize yourself with SEM fundamentals: E – K

A. GUI

The screenshot displays the VegaTC x64 SEM software interface. The main window, titled "SEM Scanning window [1024x768] 100%", shows a grayscale SEM image of a circular component with several holes. A yellow square highlights one of these holes. The interface includes a menu bar at the top, a floating toolbar on the right, and a side bar on the far right containing system status and acquisition parameters. At the bottom, an info bar displays technical details like SEM HV (5.0 kV), WD (35.86 mm), and SEM MAG (8 x). The software title "VEGA3 TESCAN" and the user's affiliation "University of California Riverside" are also visible.

Menu Bar

SEM Scanning Window

Floating Toolbar

Focus Window

Info Bar

Side Bar

SEM HV: 5.0 kV
SEM MAG: 8 x

WD: 35.86 mm
Det: SE

VEGA3 TESCAN
10 mm University of California Riverside

System status: Adjustment:

Main Toolbar

Pad

WD: 14.000 mm 0.000 mm

Info Panel

Continual Single Acquire

Scan Mode: WIDE FIELD
HV: 20.00 kV
Magnification: 22 x
View field: 12636 µm

SEM Detector

SE 35.8 % / 90.2 %

Electron Beam

HV: Off Emission: Off

Live Time: 162 h Heating: 42.5 %
HV 20.00 (10..20) kV Adjustment >>>

Vacuum

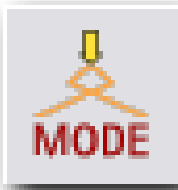
Column pressure: 6.4e+003 Pa

STANDBY VENT PUMP

Nano Stage Cont...

3

B. Floating Toolbar – 1/2



MODE: Opens the context menu for selecting *Displaying Modes*



SPEED: Opens the context menu for selecting predefined *Scan Speeds*



MAG: Left-click sets the *Magnification* as active function. Right-click opens context menu with predefined values of magnification.

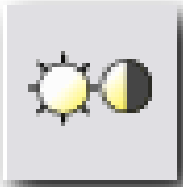


WD: Left-click sets the *Focus* as active function.



STG: Left-click sets the *Stigmator* as active function.

B. Floating Toolbar – 2/2



Brightness: Left-click sets the **Brightness and Contrast** control as active function



Auto: Left-click starts **Automatic Brightness and Contrast**



BI: Left-click sets the **Beam Intensity** as active function



Manual Column Centering: Left-click starts the manual column centering process



Acquire: Left-click starts the **Image Acquisition**

C. Sidebar – 1/5

Opens new scanning window

Opens up IR Camera

Indicates Active Function

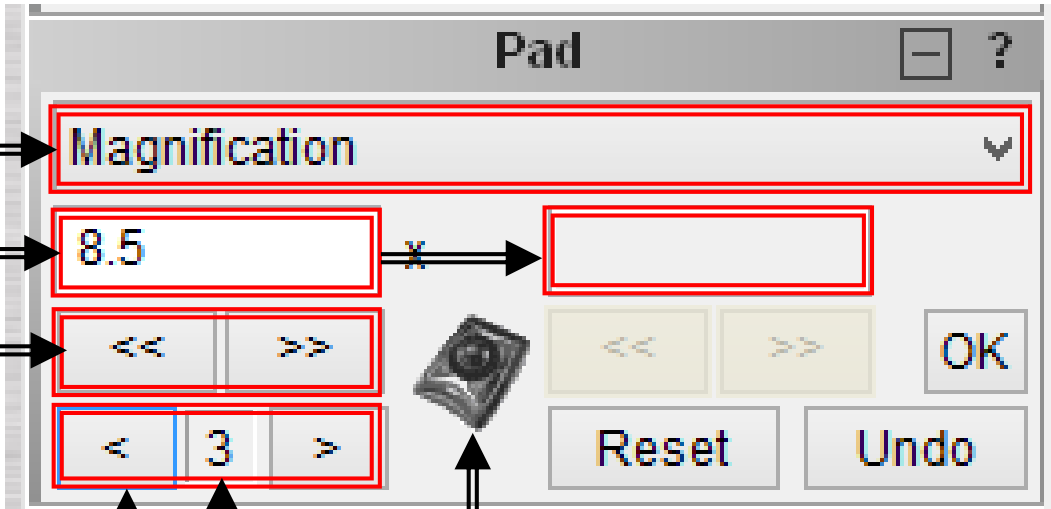
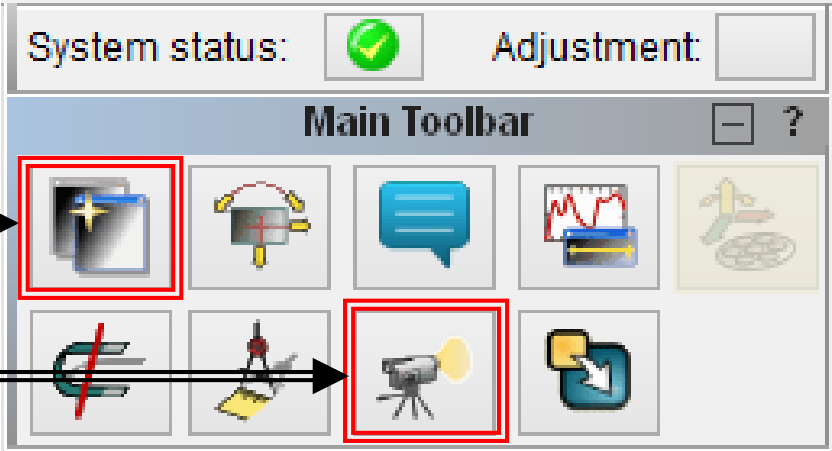
Current value(s) of Active Function

Incremental changes of Active Function

Changes Sensitivity of Increment buttons and Trackball
1 = Fine, 9 = Course

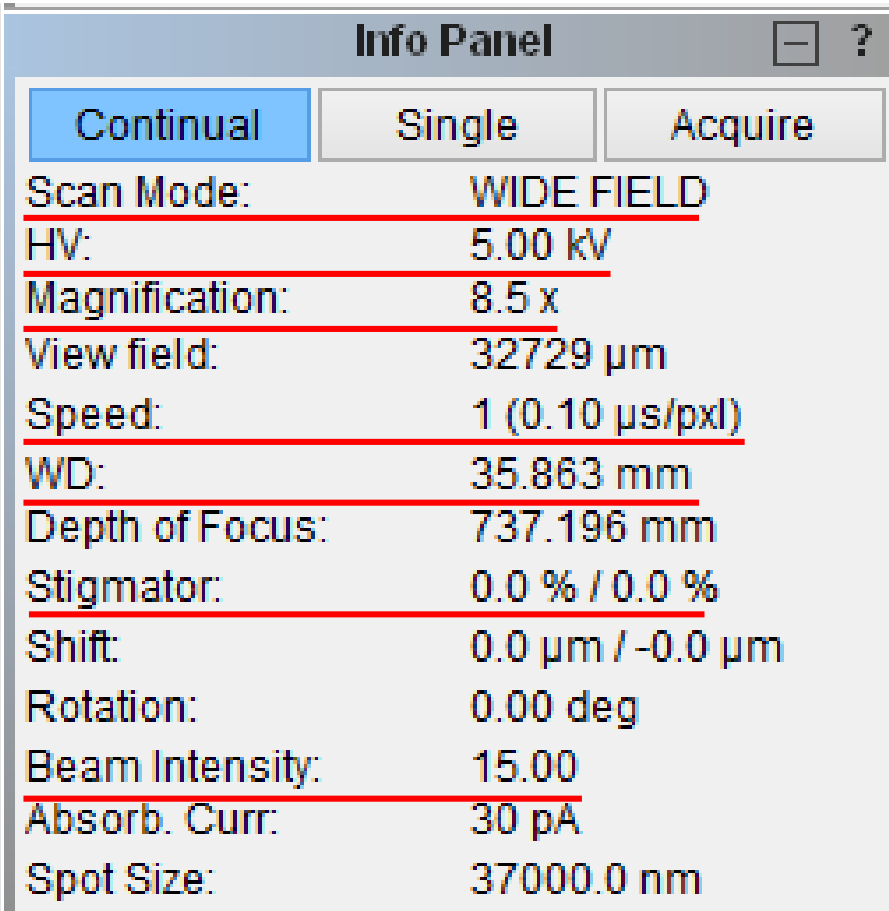
Current value of Sensitivity

Flashing Trackball Icon = Trackball is locked



C. Sidebar – 2/5

Info Panel shows all the important parameters of the microscope, and at the same time allows a quick set-up of all the most frequently used functions.



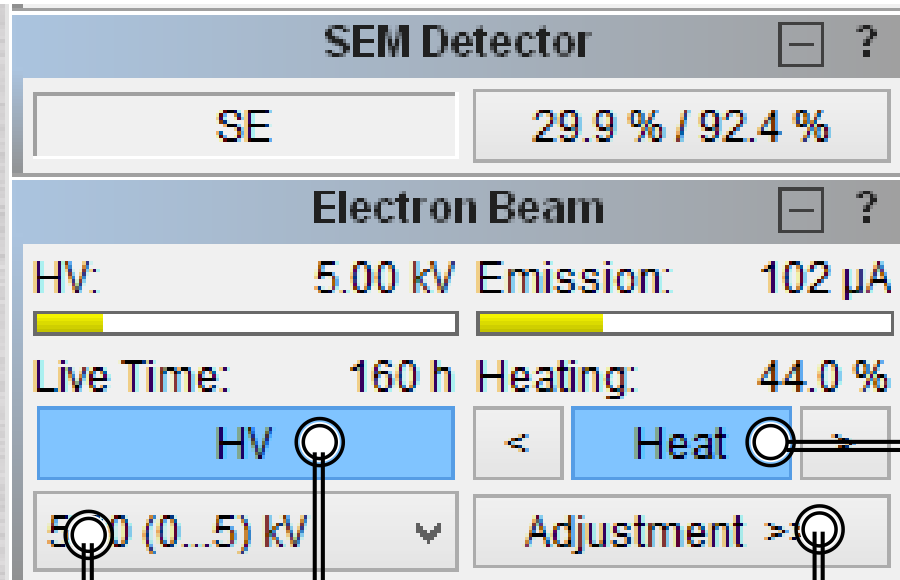
- **Continual** button stops or starts scanning
- **Single** button starts scanning of a single frame and then stops scanning
- **Acquire** button starts the acquisition process
- **HV** button sets the High Voltage value as active function
- **Depth of Focus** shows estimated range sample surface is in focus
- **Absorb. Curr.** shows the electron current absorbed by the sample
- **Spot Size** shows the sample impinging beam size

C. Sidebar – 3/5

Detector Panel shows active detector.

Electron Beam Panel controls filament heating and high voltage range.

- **SE** indicates Secondary Electron detector is active
 - %/% shows Brightness/Contrast
- **HV** shows high voltage value
- **Emission** shows current emitted
- **Live Time** shows total working time of filament
- **Heating** shows relative value of filament heating current in %



Heat starts or stops filament heating

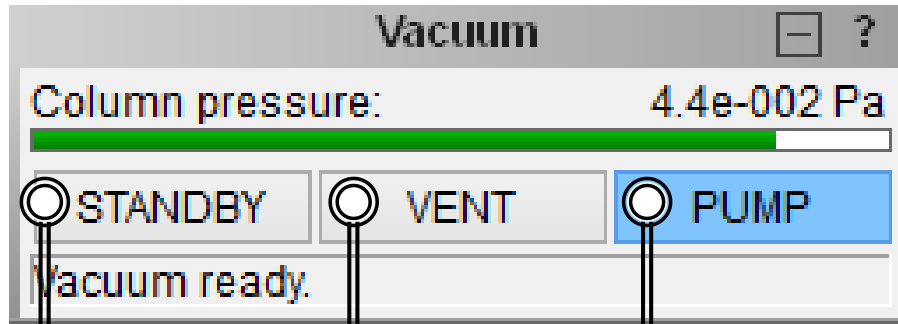
Adjustment opens context menu

HV turns the high voltage on and off

HV Drop Down selects HV range

C. Sidebar – 4/5

Vacuum Panel controls the vacuum system.



PUMP starts the pumping procedure (~ 3 min to complete)

VENT vents the microscope

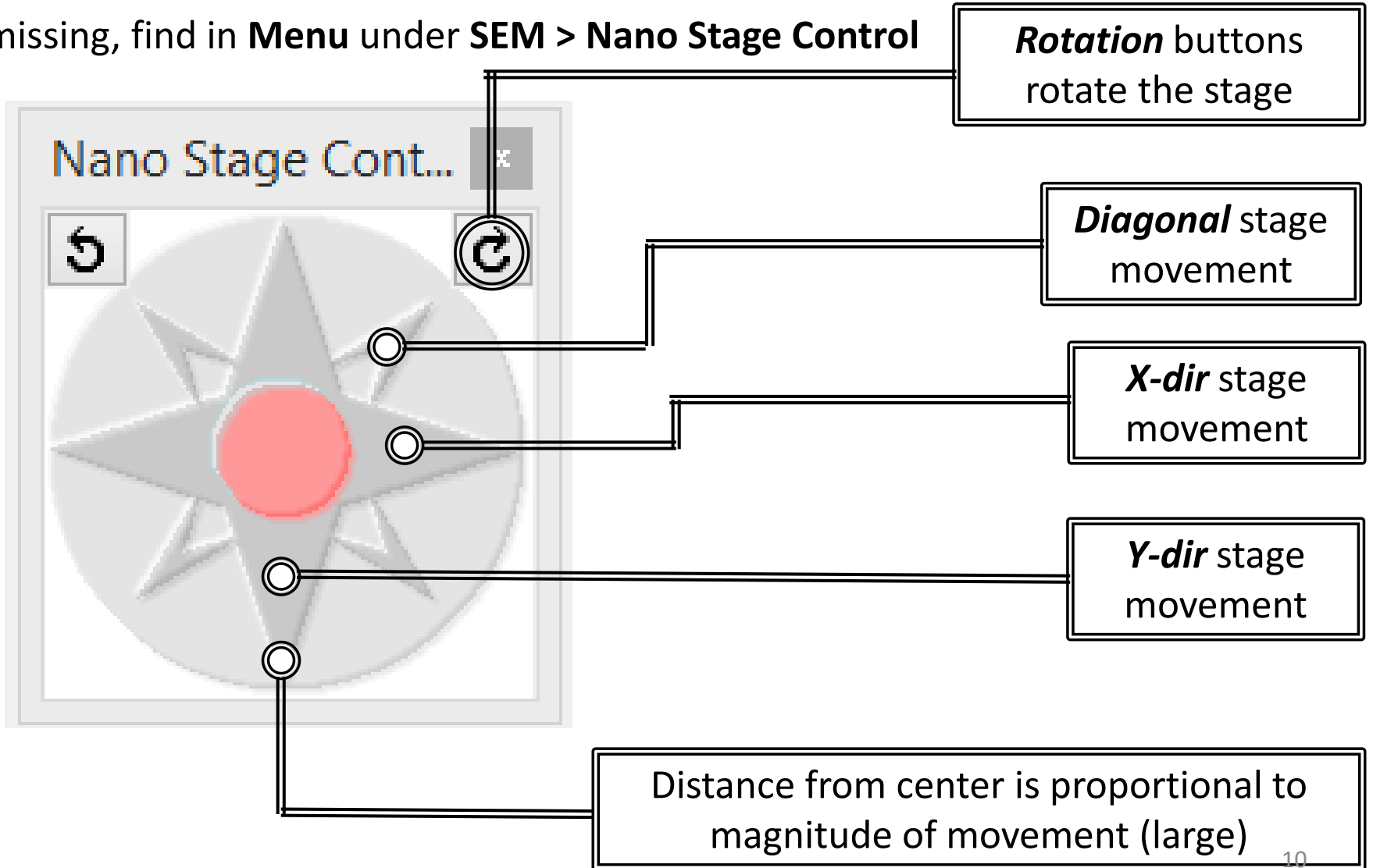
STANDBY interrupts microscope work if necessary for a long period of time

- **Column Pressure** indicates the value of the pressure in the column
 - Red = Not Ready
 - Green = Ready
- **Status** shows state of vacuum
 - **Venting** = still venting
 - **Venting finished** = venting is finished and chamber can be opened
 - **Pumping** = still pumping
 - **Vacuum ready** = chamber is pumped down to sufficient vacuum
 - **Vacuum off** = vacuum is in standby mode

C. Sidebar – 5/5

Nano Stage Control controls the specimen stage movement.

If missing, find in **Menu** under **SEM > Nano Stage Control**



D. SEM Image Parameters

Windows

Aspect Ratio of Image = 4:3

Resolution = 1024 x 768

Averaging

Accumulation = Disable

(Prone to vibrational noise and drift)

Acquisition

Keep Actual Speed

Keep view field/print magnification

Infobar Texts

Show Infobar:

Beam Energy, Working Distance, View Field,
Detector, Vacuum, Scan Mode

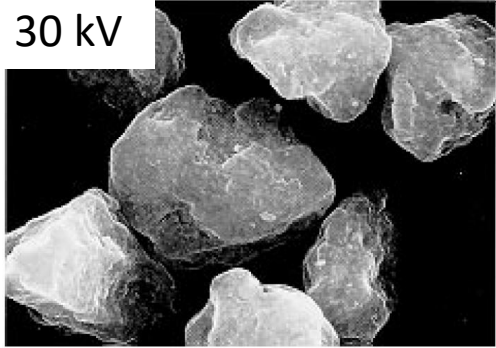
The screenshot shows the 'SEM Image Parameters' dialog box with the following settings:

- Windows:** Live: 4:3, 1024 x 768; Save: 4:3, 1024 x 768.
- Averaging:** Enable button; Live: Frame accumulation, 5; Save: Frame accumulation, 5.
- Acquisition:** Acquisition time: 0.12 s; Keep actual speed; Keep view field / print magnification.
- Infobar texts:** Show infobar checked; Beam energy, Working distance, Magnification, Detector, View field, Date; Burn Note & Sign texts into image unchecked.

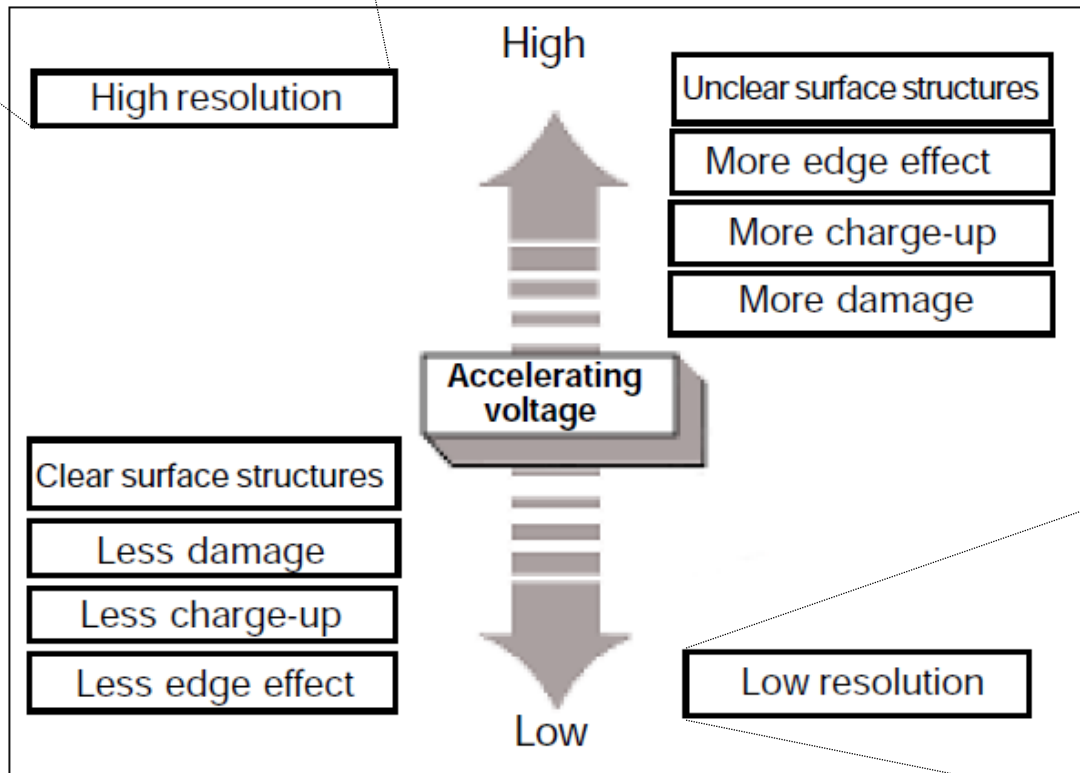
An 'Apply' button is located at the bottom right of the dialog box.

E. Accelerating Voltage – 1/2

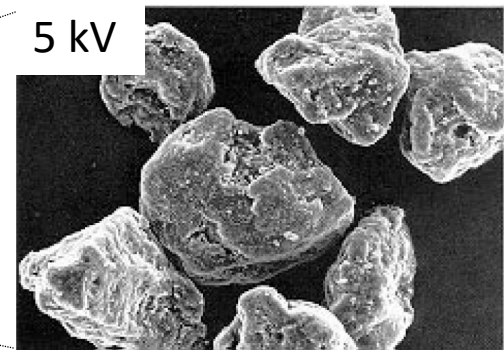
30 kV



Recommendation: Start at **5 kV** and increase voltage incrementally to balance resolution to surface structures

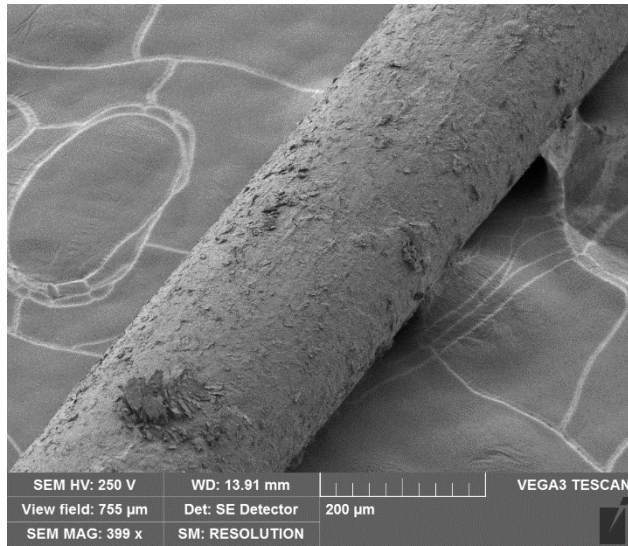


5 kV

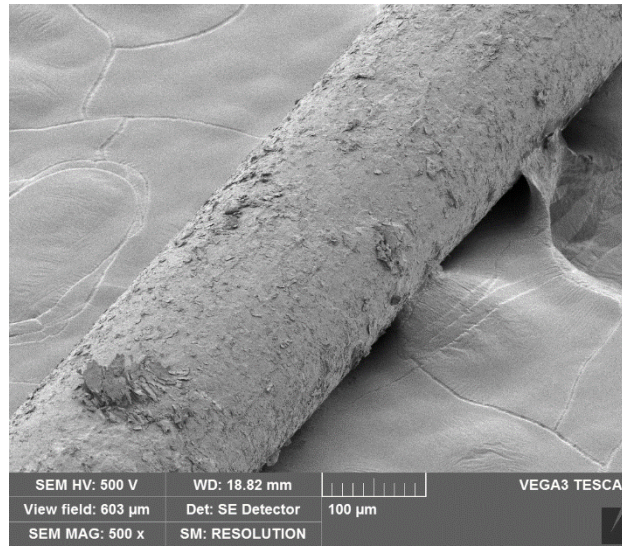


E. Accelerating Voltage – 2/2

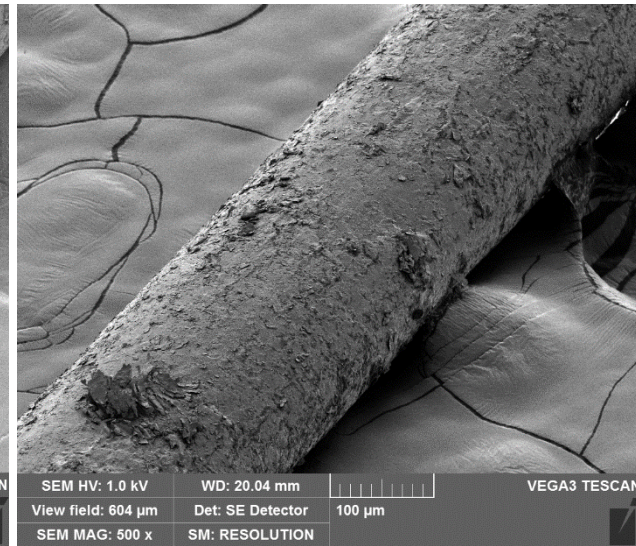
250 V



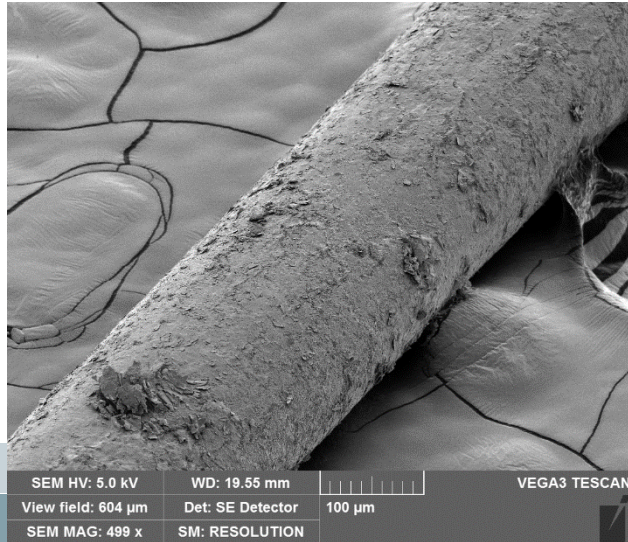
500 V



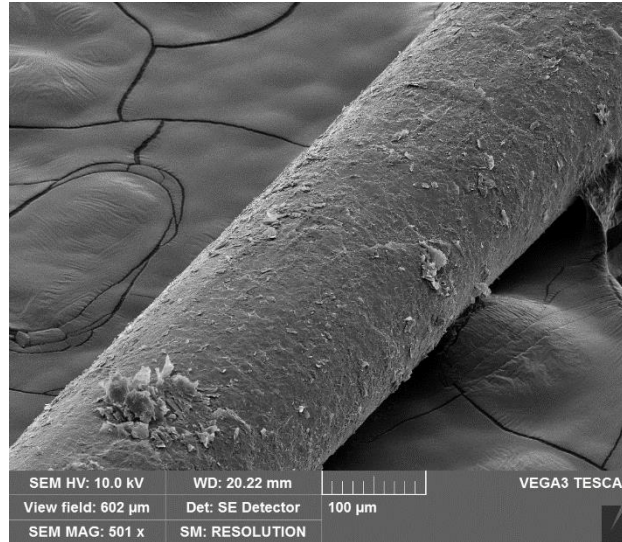
1 kV



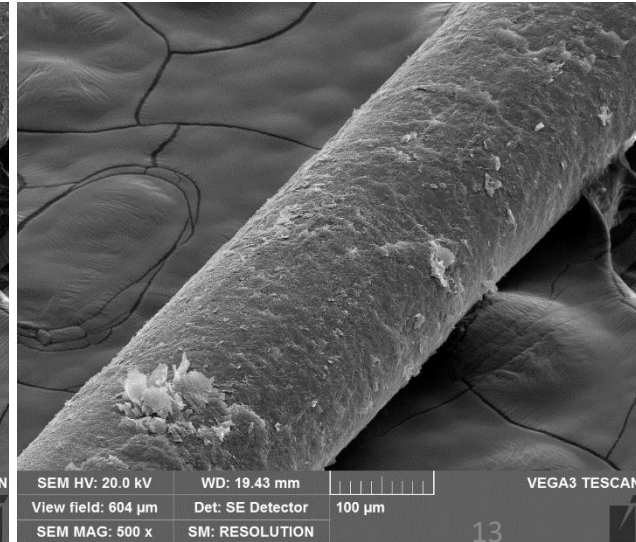
5 kV



10 kV

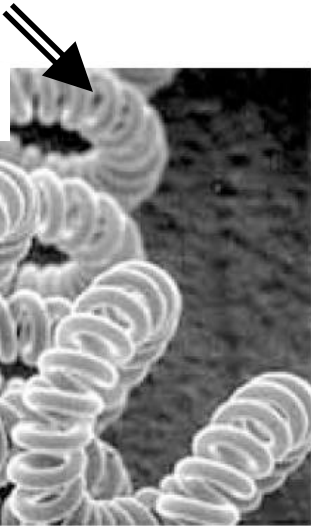


20 kV

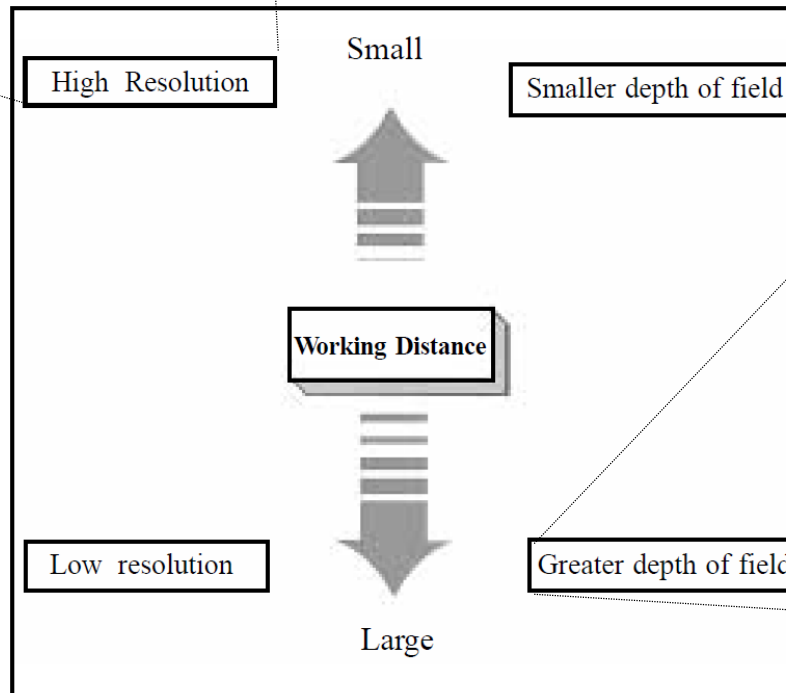


F. Working Distance

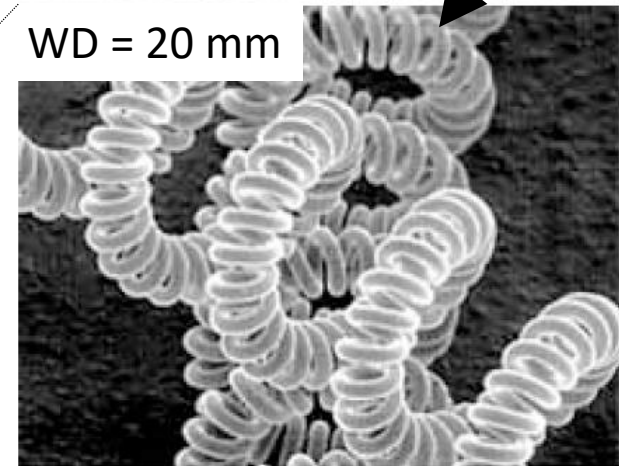
WD = 10 mm



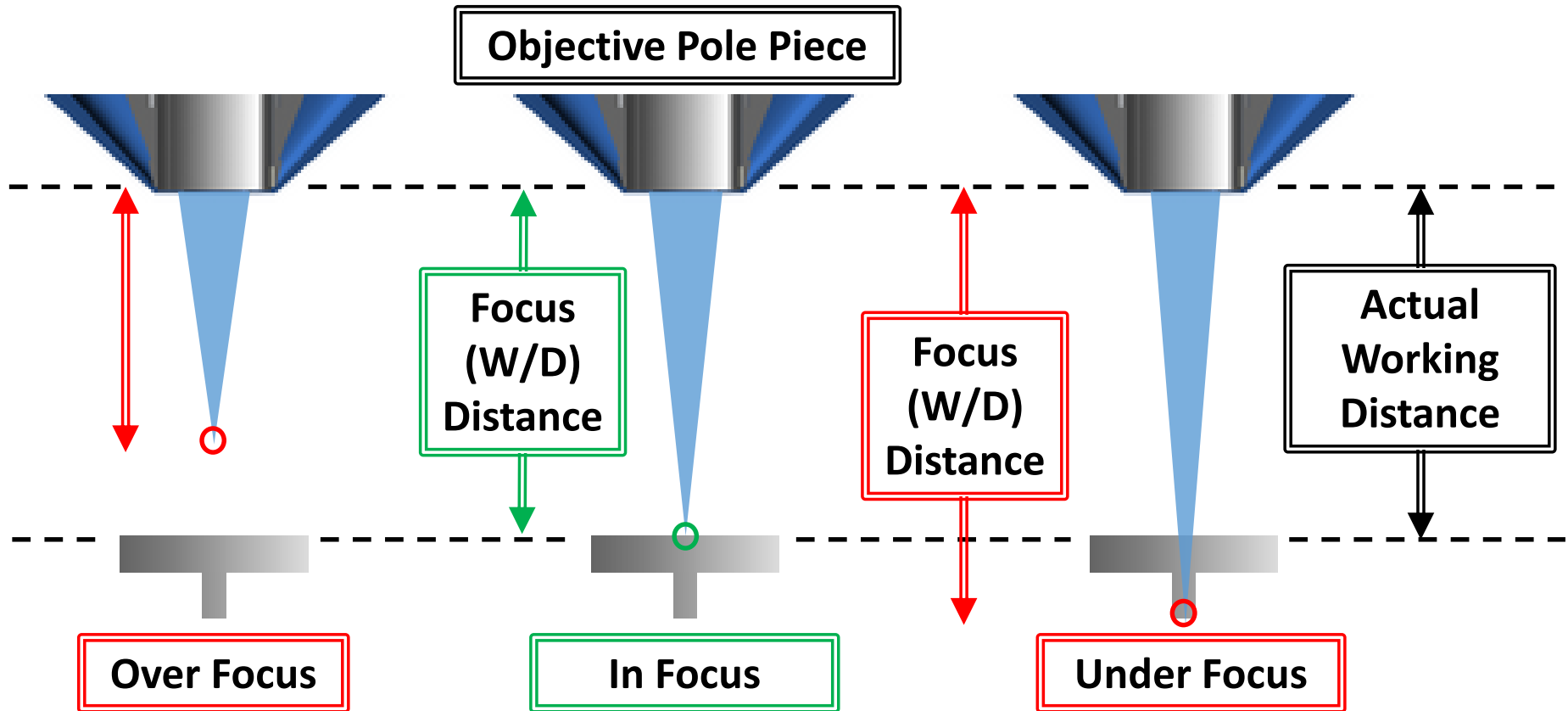
Recommendation: Start at ≈ 10 mm and decrease WD to achieve greater resolution or increase WD to achieve greater depth of field if necessary



WD = 20 mm



G. Working Distance vs Focus (W/D) Distance

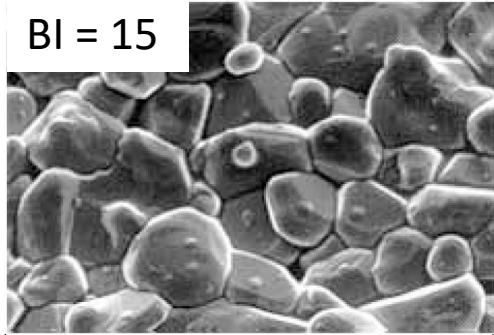


Actual Working Distance = Distance between objective pole piece and sample and can only be controlled manually with the **knob outside the chamber**

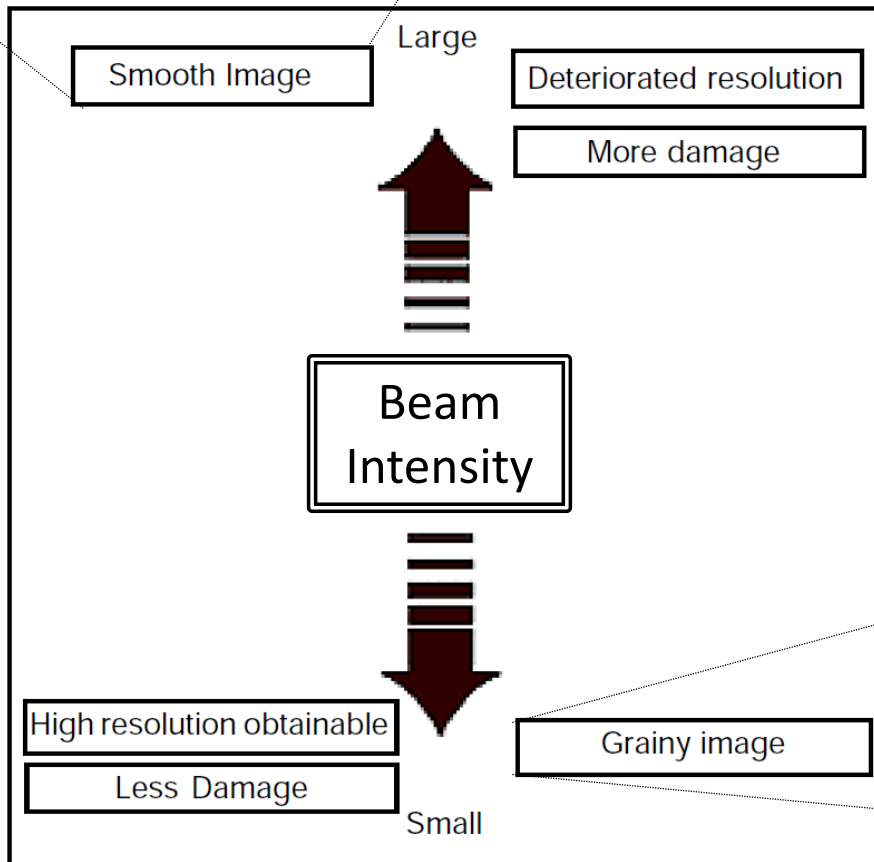
Focus (W/D) Distance = Distance between objective pole piece and focal point and can only be controlled by the **Focus (W/D) button**

H. Beam Intensity – 1/2

BI = 15



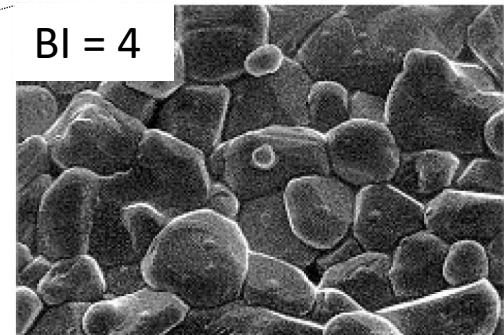
Recommendation: Decrease beam intensity until balance between resolution/grainy image and acquisition time is desired



High BI: Larger spot size for low magnification but poor resolution

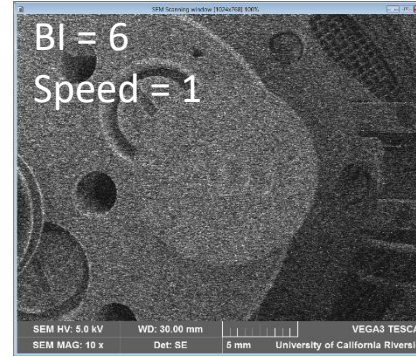
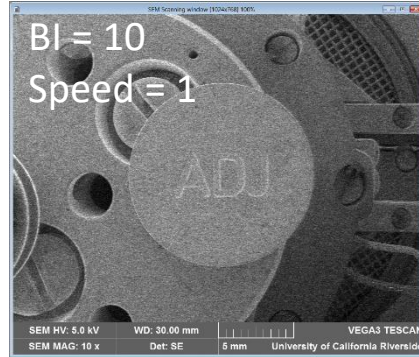
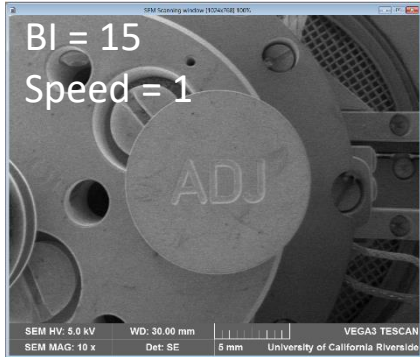
Low BI: Higher resolution but grainier image, need to balance with slower scan **SPEED**

BI = 4



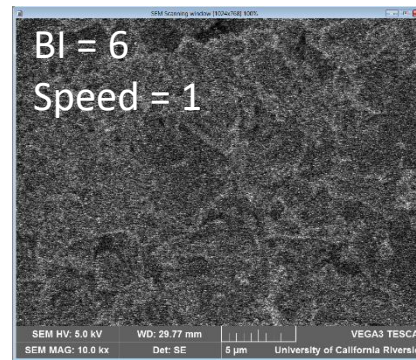
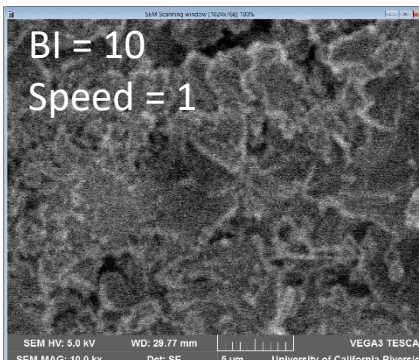
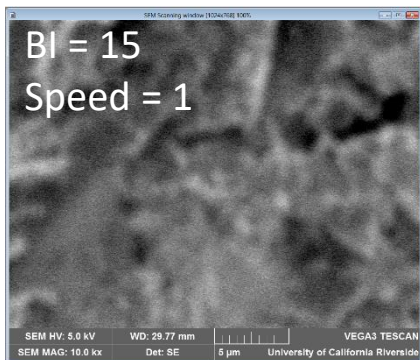
H. Beam Intensity – 2/2

Low Magnification (Minimum < Mag < 10 kX)



At Low Mag, lowering BI doesn't have a dramatic affect on the quality of image...

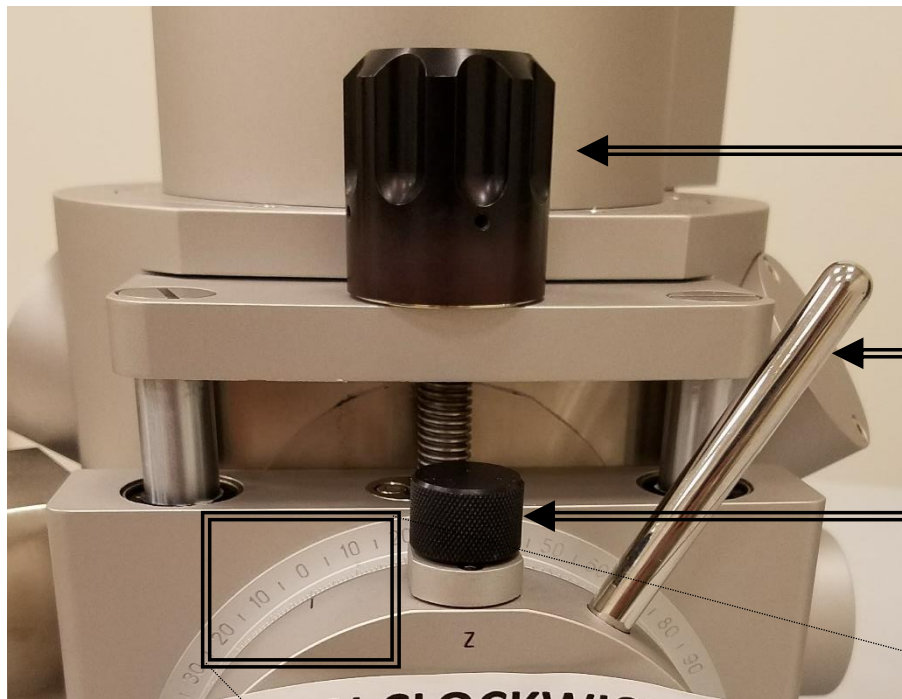
High Magnification (10 kX < Mag < Maximum)



At High Mag, the BI **MUST** be chosen correctly!

A grainy image will **ALWAYS** accompany a reduction in BI, but is easily removed with
a drop in scan **SPEED!**

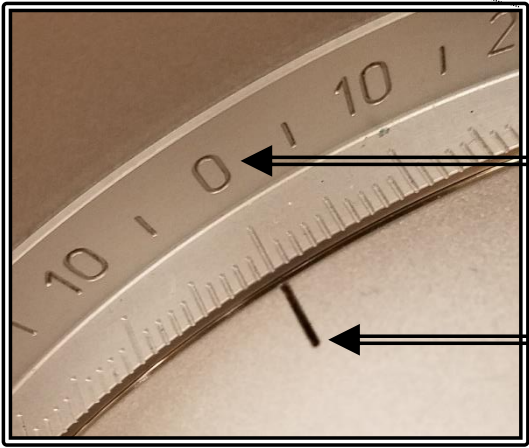
I. SEM Chamber



Z-dir Stage Movement Knob – Coarse Control

Stage Tilt Handle

Z-dir Stage Movement Knob – Fine Control



0° Tilt Position

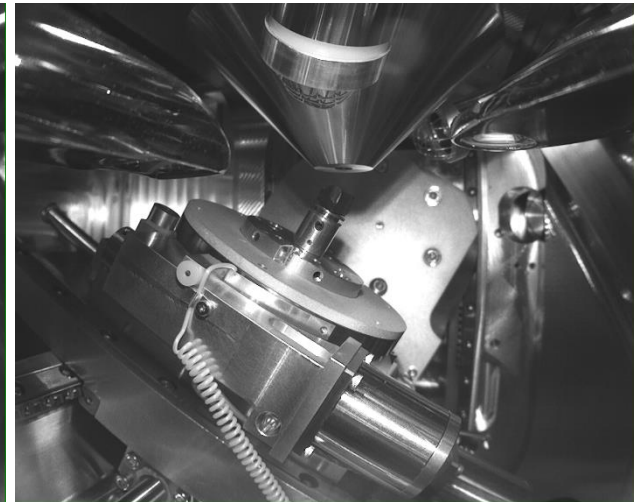
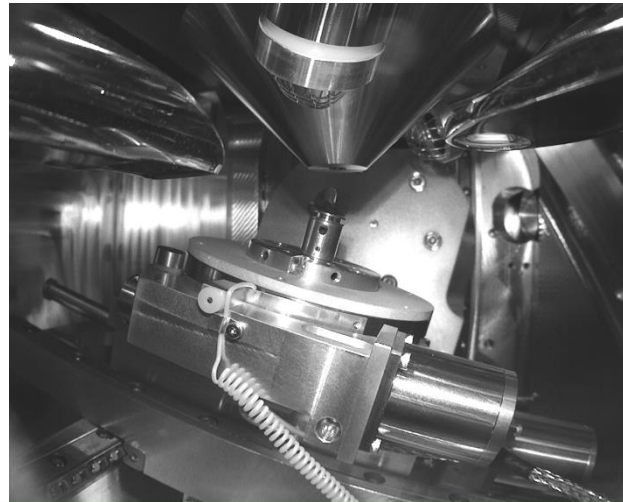
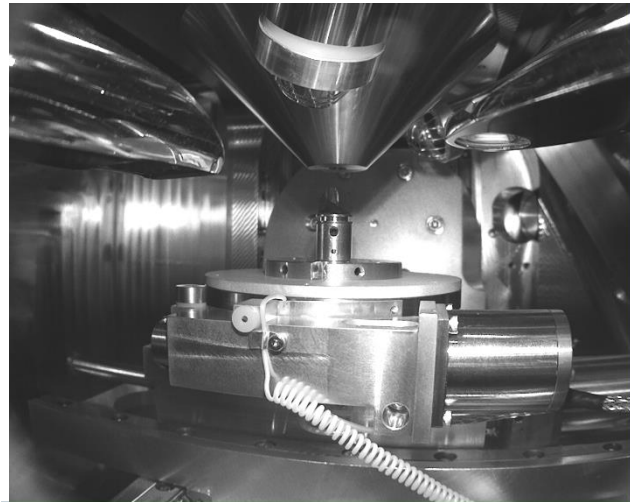
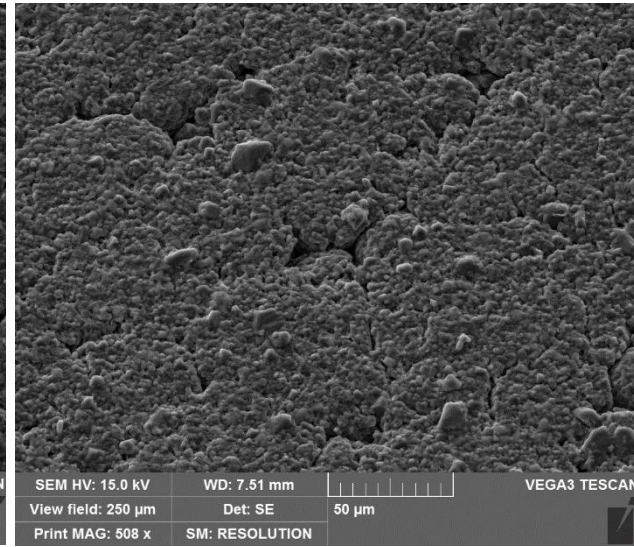
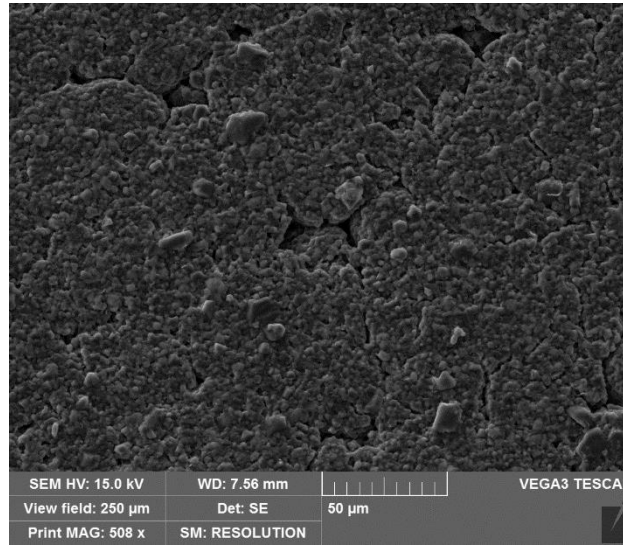
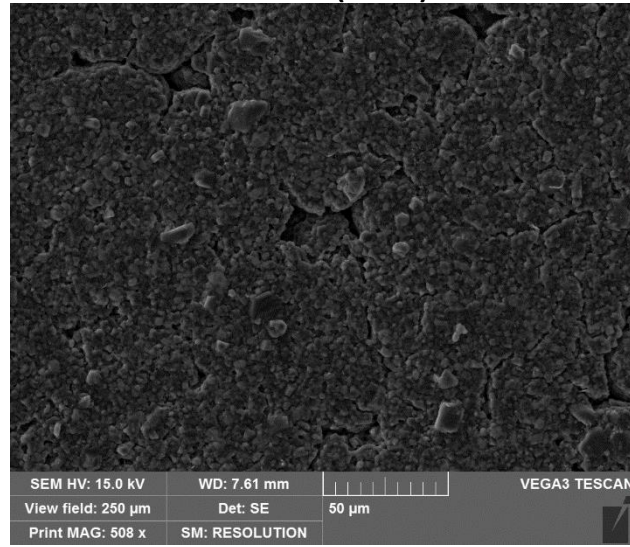
Tilt Position Marker

J. Tilt (Advanced Users) - 1/2

0° Tilt (Flat)

15° Tilt

30° Tilt

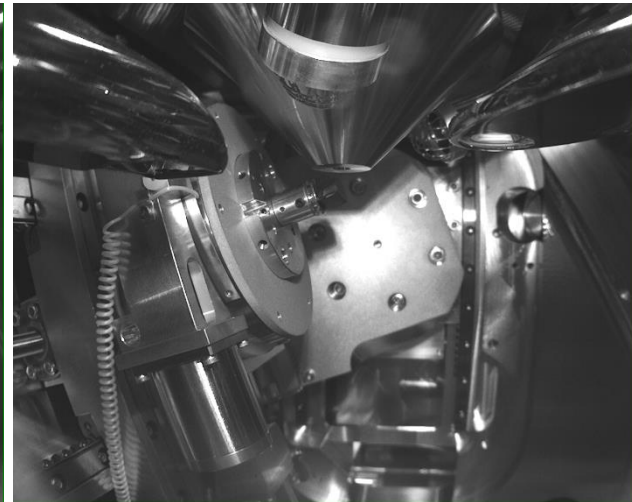
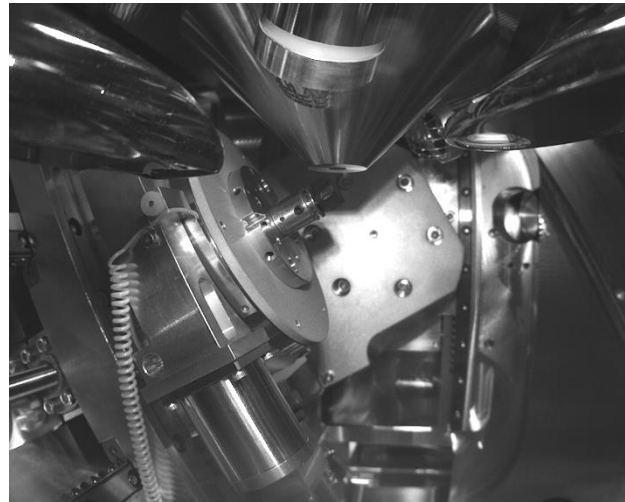
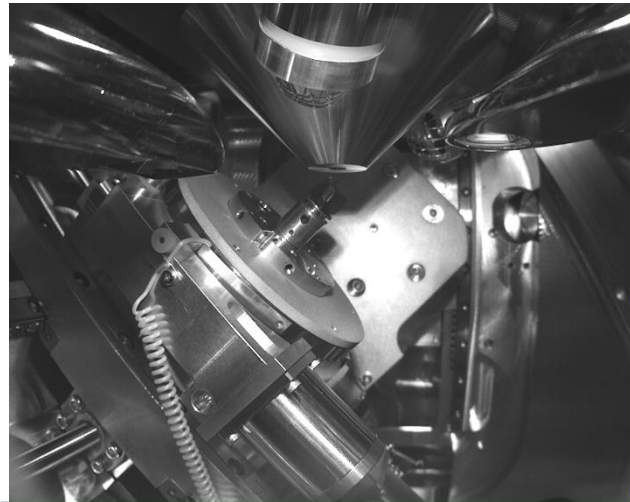
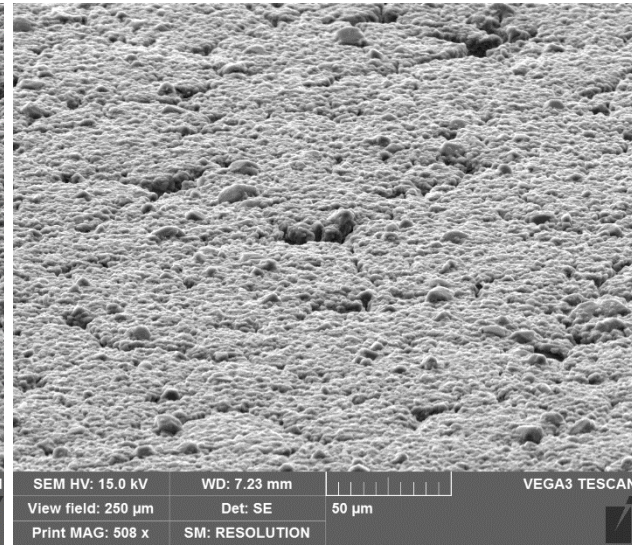
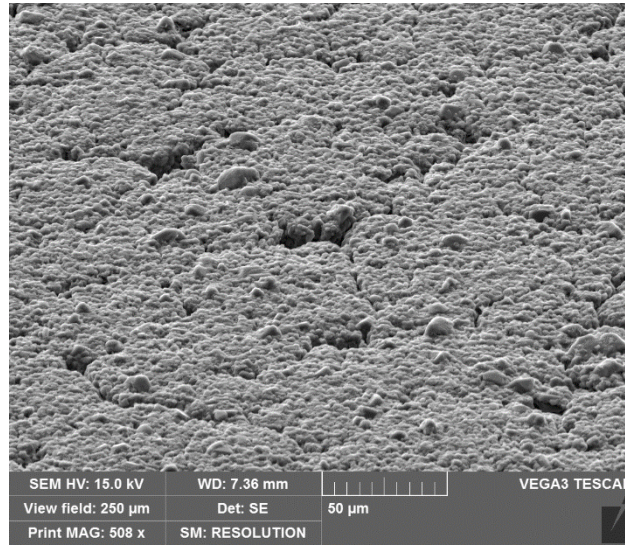
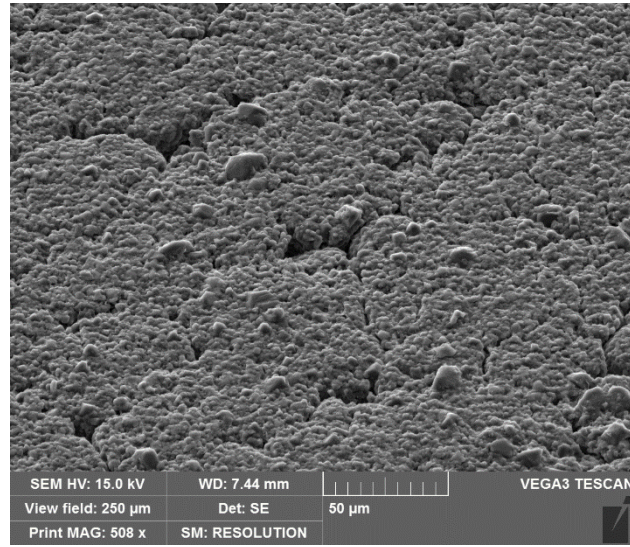


J. Tilt (Advanced Users) - 2/2

45° Tilt

60° Tilt

70° Tilt



K. High Resolution Imaging Process Tree

#	Description	Stage	Mag	Focus	Z Knob	BI	Speed	Auto B/C
1	Center tallest part of tallest sample in window	Yes	Yes	Yes		Yes	Yes	Yes
2	Achieve desired working distance			Yes	Yes	Yes	Yes	Yes
3	Center desired sample image in window with desired Mag	Yes	Yes	Yes		Yes	Yes	Yes
4	Increase Mag to \geq 2X desired Mag		Yes	Yes		Yes	Yes	Yes
5	Beam optimization (if desired Mag \geq 10 kX)			Yes		Yes	Yes	Yes
6	Achieve best focus			Yes		Yes	Yes	Yes
7	Reduce Mag back to desired Mag		Yes			Yes	Yes	Yes
8	Determine optimal image conditions for BI and Speed and acquire					Yes	Yes	Yes
9	Reduce Mag and acquire image		Yes			Yes	Yes	Yes
10	Move to new sample location -> Repeat #3 to #9							

SEM Operation

- I. Initiate Software
- II. Sample Preparation
- III. Sample Loading
- IV. Turning on HV
- V. Mode
- VI. Beam Intensity
- VII. Brightness and Contrast
- VIII. Mag
- IX. Focusing
- X. Speed
- XI. Working Distance
- XII. Image Preparation
- XIII. Column Centering
- XIV. Stigmation Correction
- XV. Image Acquisition
- XVI. Saving
- XVII. Sample Unloading
- XVIII. Cleanup

I. Initiate Software – 1/1

1. **Record** your time-in on the **sign-in sheet** located on preparation table
2. Sign into Windows using provided **Username** and **Password** located on **monitor** if necessary

3. Double-click on VegaTC icon to load software



4. Sign into your user account with your **Username** and **Password**



II. Sample Preparation – 1/2

1. **Always wear gloves** when dealing with anything that will be placed into or in contact with the SEM
2. The specimen should be conductively fixed with double-sided carbon tape to a specimen stub (12.5 mm specimen pin-stubs)



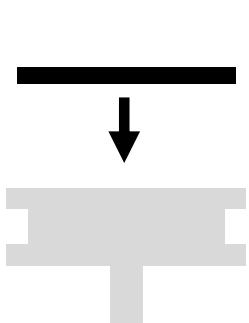
Double-sided carbon tape (DCT)



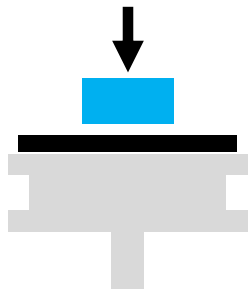
SEM specimen stub

3. For non-conductive samples, “sandwich” the carbon tape around your specimen to provide a conductive pathway

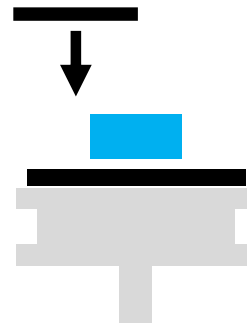
Place initial DCT on stub



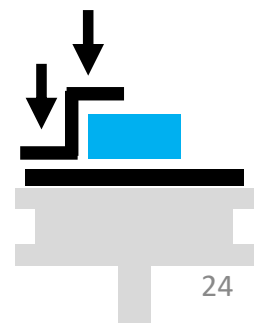
Place specimen and press down



Place top DCT over sample and initial DCT



Press down on the top DCT to ensure contact



II. Sample Preparation – 2/2

4. Follow the steps for operating the Sputter Coater (requires training) to deposit a thin-layer of gold



5. Magnetic samples will need to be fixed well by a screw holder (provided by user)

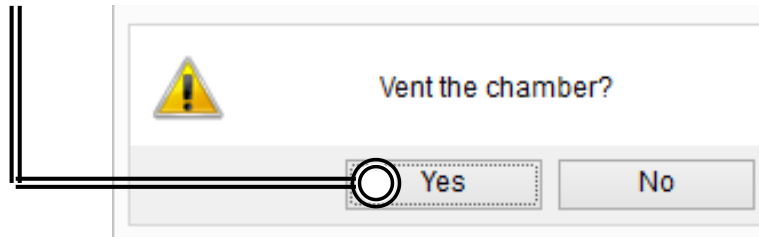
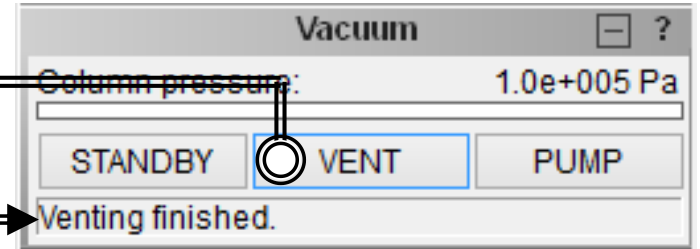


6. Items located in the cabinet are available for SEM users to help prepare their samples

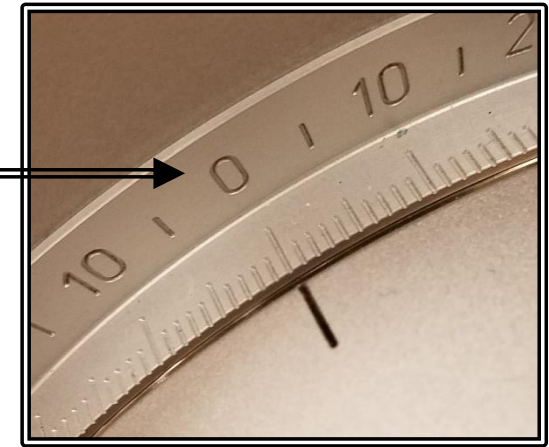


III. Sample Loading – 1/4

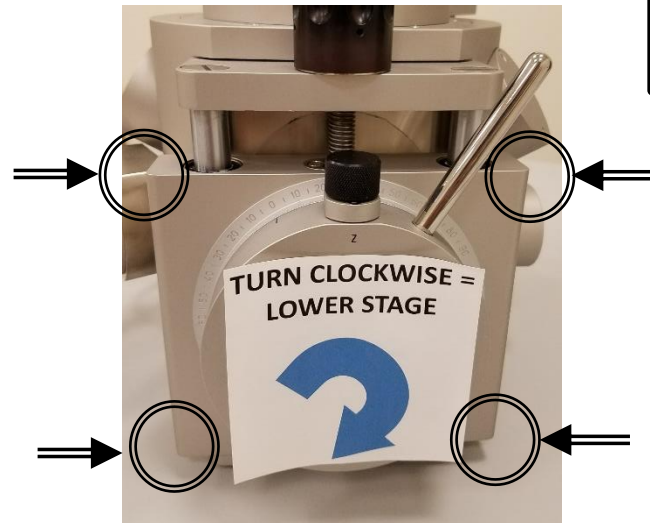
1. Click **VENT** to vent the microscope
2. Click **Yes** to confirm venting



3. Wait until "**Venting finished**" appears
4. Set the tilt of the specimen stage to 0° if not already set to 0° (**Advanced Users only**)

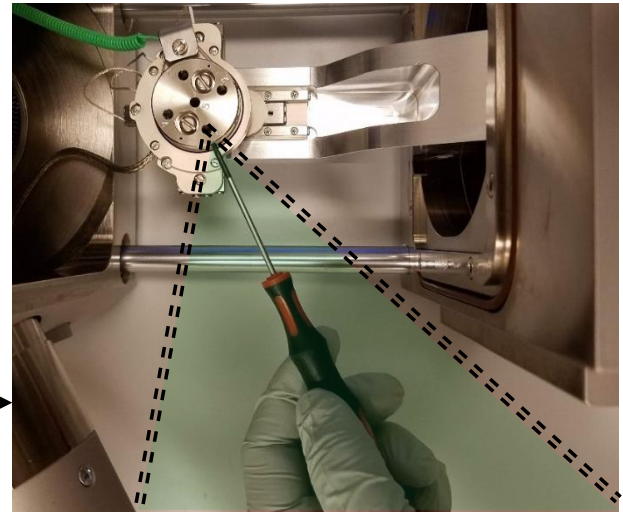


5. Gently pull the chamber corners toward you to open the chamber



III. Sample Loading – 2/4

6. Rotate stage if necessary to access screw port in preferred orientation for maximum clearance



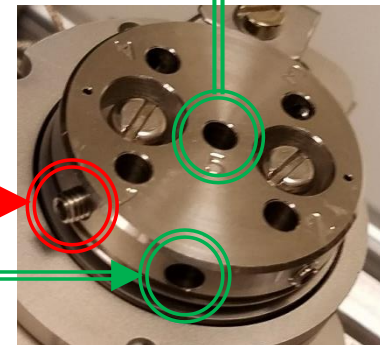
7. Using provided tweezers, clamp onto the specimen stub and **blow a stream of air** over the **entire specimen stub** AWAY from the chamber using Airgun



8. Loosen the screw first (see example)

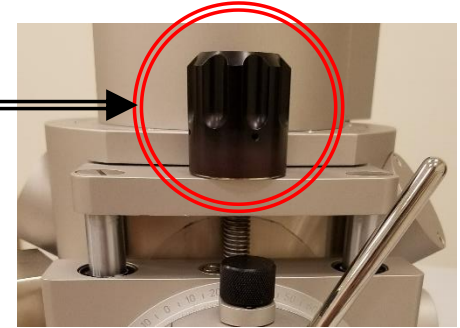
9. Carefully insert the specimen stub into the specimen stage

10. Tighten the screw holding the specimen stub



III. Sample Loading – 3/4

11. Ensure that the sample stage is at the lowest position using Z-knob (clockwise)

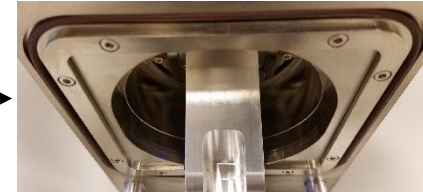


12. Check to see if the O-ring along the chamber door is snugly sitting inside the recessed groove

Not Snug!



Snug



13. If loose, WEARING CLEAN GLOVES run your finger along the O-ring to ensure that it is sitting inside the recessed groove



14. Carefully close the chamber door by pushing it towards the chamber

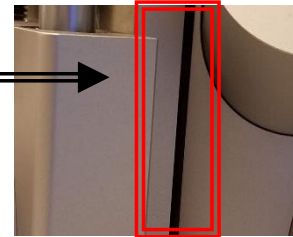
CHECKING THAT THE SAMPLE DOES NOT TOUCH ANYTHING INSIDE CHAMBER



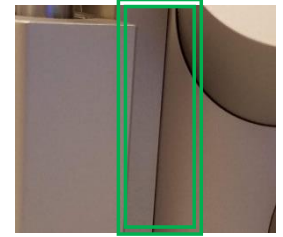
III. Sample Loading – 4/4

15. Ensure there is no gap between chamber and door, else fix O-ring following steps 12 and 13 again

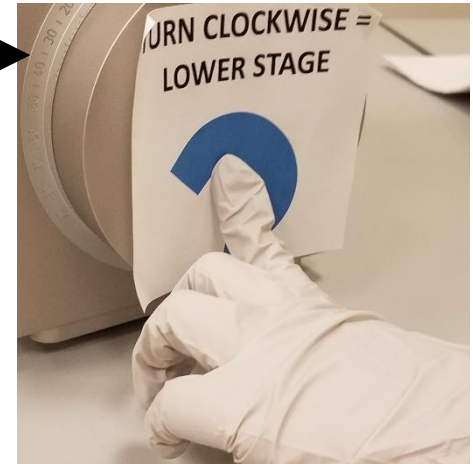
Gap



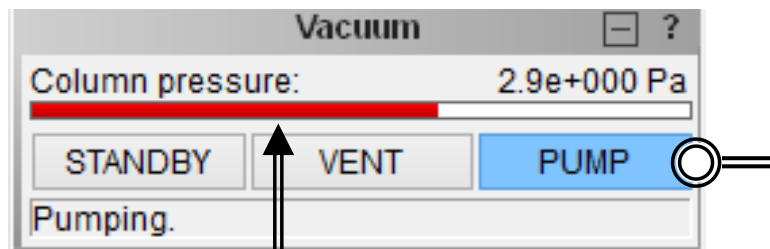
No Gap



16. Place finger against chamber door

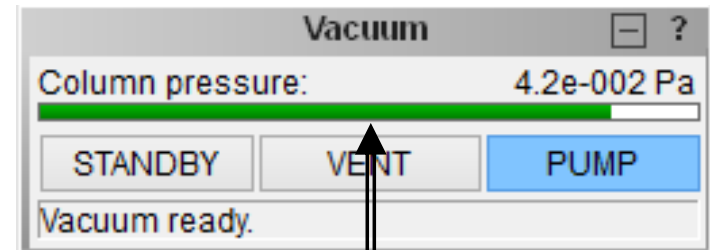


17. Click **PUMP** to start pumping down chamber



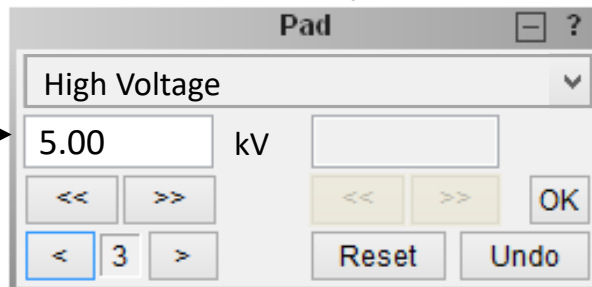
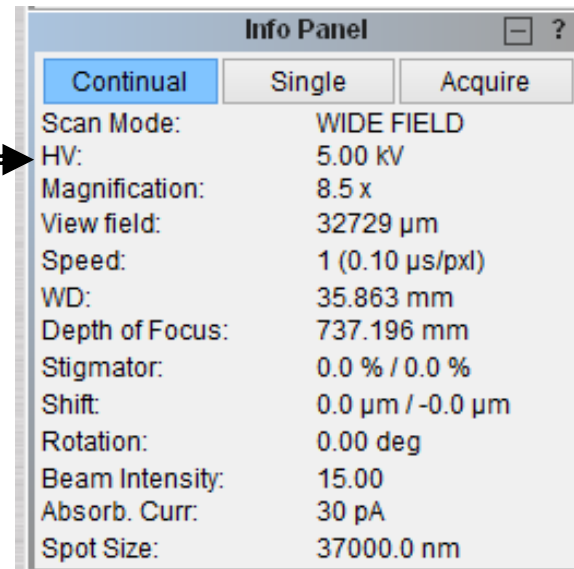
18. Wait until bar graph shows red to release finger

19. Wait until the bar graph turns green or "**Vacuum ready**" appears (~ 3 min)



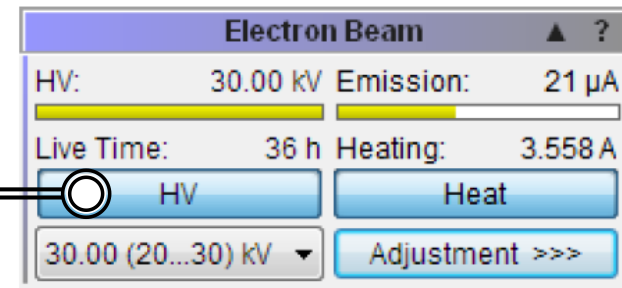
IV. Turning on HV – 1/1

1. Click on **HV** on the Info Panel or select **HV** in Pad Drop Down

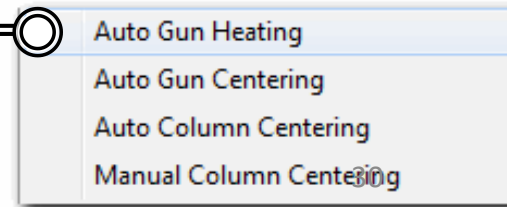
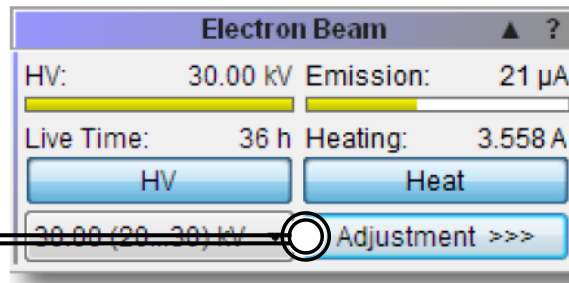


2. Set a specific **High Voltage** in the **Pad** panel (set 5 kV as starting voltage)

3. Click **HV** to turn on the high voltage

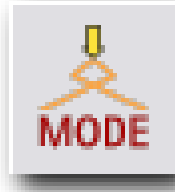


4. Click **Adjustment >>>** and select **Auto Gun Heating**



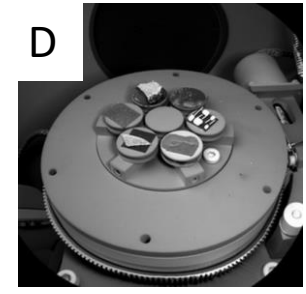
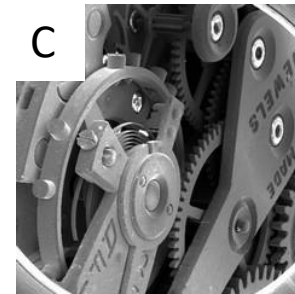
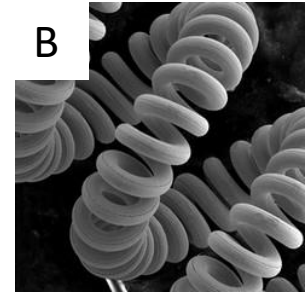
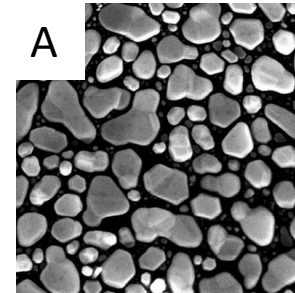
(required if black screen present AFTER turning on HV)

V. Mode – 1/1

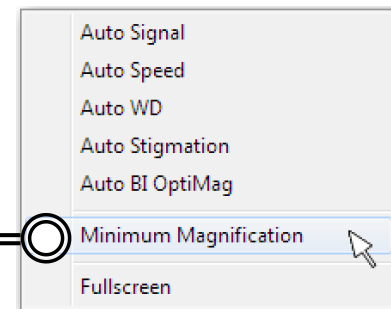


1. Click **MODE**
2. Confirm **Continual Wide Field** option is checked ✓
3. Choose desired scanning mode
(default = Resolution)

Mode	Characteristics
A Resolution	High resolution Lower depth of focus
B Depth	Good resolution Increased depth of focus
C Field	Lower resolution Large field of view High depth of focus
D Wide Field	Extra large field of view

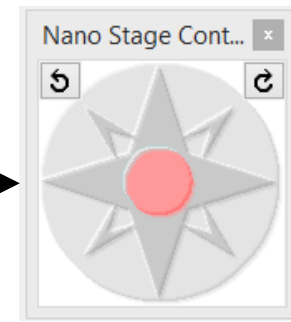


4. Right-click on **MAG** and select **Minimum Magnification**



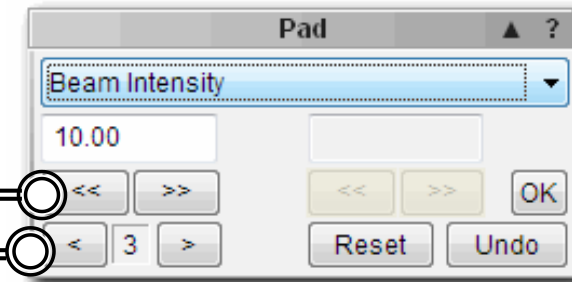
VI. Beam Intensity – 1/1

1. Center the SEM window onto your desired sample using the stage control
2. Click **BI** to adjust beam intensity using the << and >>



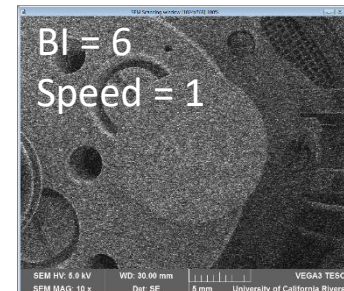
Recommended Initial **BI** values

Magnification	Beam Intensity
Min → 200	18 → 13
200 → 2000	12 → 8
2000 → 10k	10 → 7
10k → Max	7 → 4



3. Recommend **BI** of **15** to start at low mag
4. Change the sensitivity if necessary

Recommended Value = 3

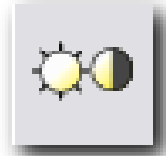


VII. Brightness and Contrast – 1/1

1. Click **Auto** to auto adjust the brightness and contrast if too bright or dark as necessary



2. Click **Brightness** to manually adjust the brightness and contrast



Contrast: Hold F12 +  trackball

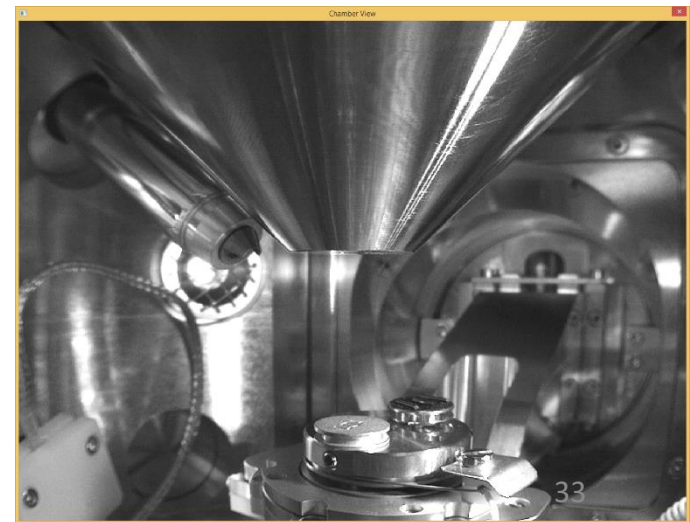
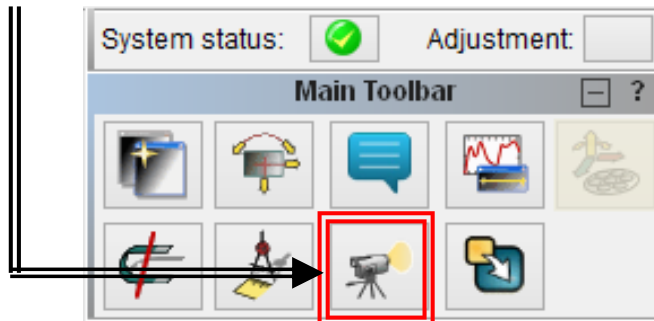


= Change only Contrast

Brightness: Hold F11 +  trackball

= Change only Brightness

3. Click on the **IR Camera** button to open up the view of the chamber (if you haven't already)

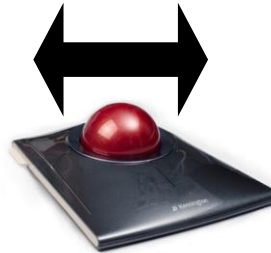


VIII. Mag – 1/1

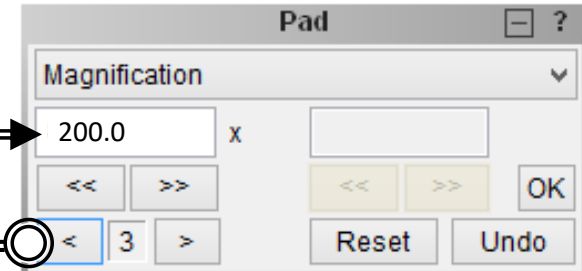
1. Click **MAG** to change the magnification



2. Turn the trackball from left to right



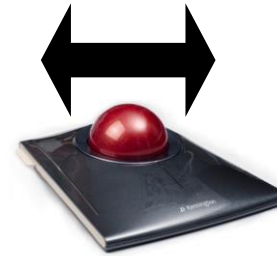
3. Or enter a value directly in **Pad** panel



4. Change the sensitivity if necessary

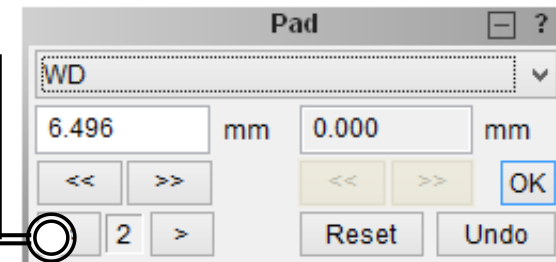
Recommended Value = 5

IX. Focusing – 1/1



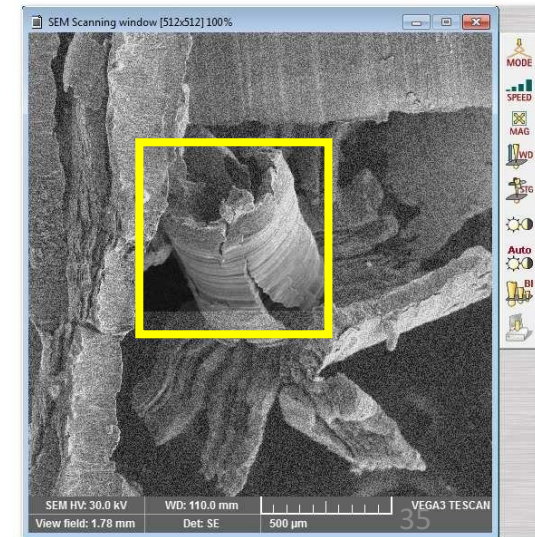
1. Click **WD** to adjust **focus distance**
2. Turn the Trackball from left to right to adjust focus
3. A **focused image** shows the **actual working distance** via **WD value**
4. Change the sensitivity if necessary

Recommended Value = 2 for Fine ($\text{Mag} \geq 10\text{kX}$)
and 5 for Coarse ($\text{Mag} \leq 10\text{kX}$)



5. Double-left-click in the SEM scanning window to create a **Focus Window**
 - Left mouse button inside = move **Focus Window**
 - Right mouse button inside = resize **Focus Window**
 - Double-left-click = remove **Focus Window**

6. $\text{WD} \approx 30 \text{ mm}$ when sample is at lowest position



X. Speed – 1/1

1. Click **SPEED** to adjust scan speed
2. Use Focus Window to determine the effect of **SPEED** and **BI** has on your image quality



SPEED	Acquisition Time
1	0.12 sec
2	0.30 sec
3	0.87 sec
4	3 sec
5	16 sec
6	32 sec
7	1 min 36 sec
8	4 min 34 sec
9	13 min 58 sec
10	44 min 4 sec

Recommendation:

SPEED of 1 – 4 for **initial focusing**

BI setting should be appropriate to **MAG** value

SPEED of higher values looks better but takes longer to focus!

Use higher **SPEED** values of 5 – 8 when **ready to save images**

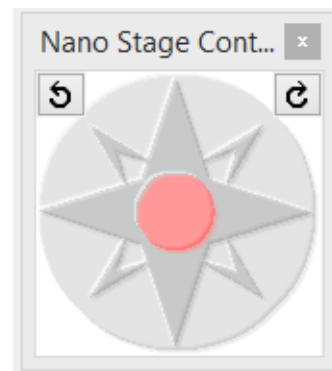
Recommended Initial **BI** values

Magnification	Beam Intensity
Min → 200	18 → 13
200 → 2000	12 → 8
2000 → 10k	10 → 7
10k → Max	7 → 4

XI. Working Distance – 1/3

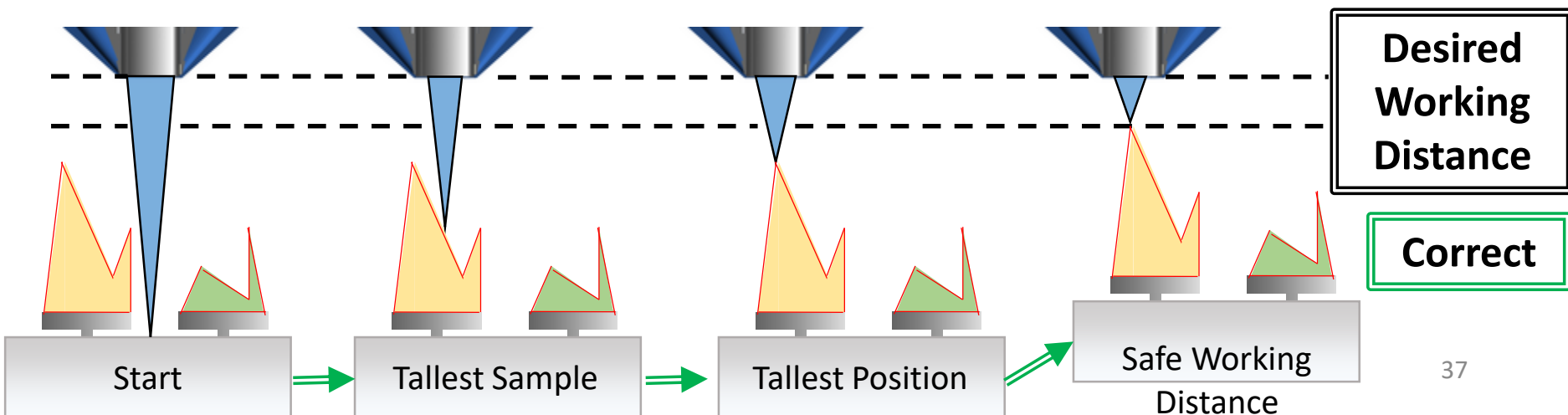
Use combination of **MAG**, **Stage Control**, and focusing (**WD**)

- Identify and bring the **tallest position** of your **tallest sample** to the **center** of SEM scanning window
- Increase **MAG** until **distinct features** make up **majority** of window
- Check if mode = **Resolution** or **Depth** (if not, keep increasing **MAG**)
- If you can't see transition between focus & out-of-focus with **WD**, you **skipped a step!**



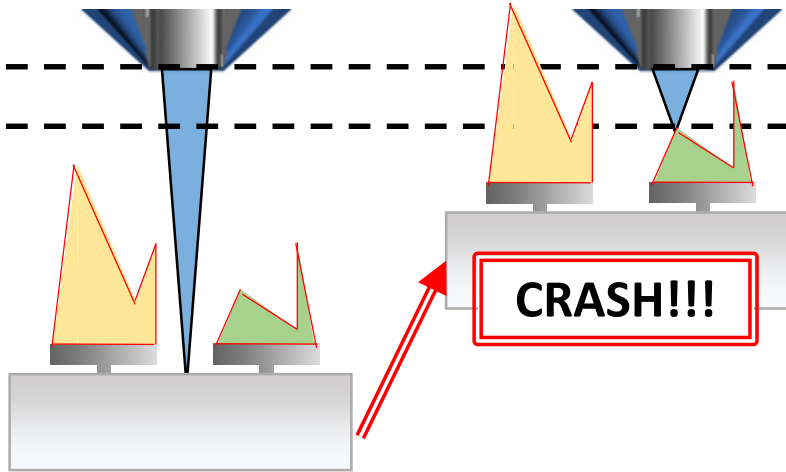
NOTE: The tallest portion of the tallest sample should be focused since this will crash into the pole-piece first as you raise the stage in the next step.

This **DOES NOT** have to be the desired position or sample for your images, it is **ONLY** for setting the safe working distance value!

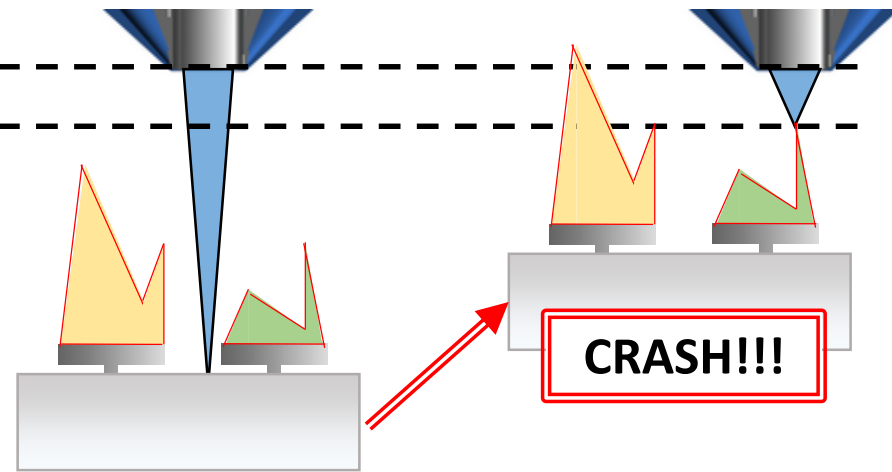


XI. Working Distance – 2/3

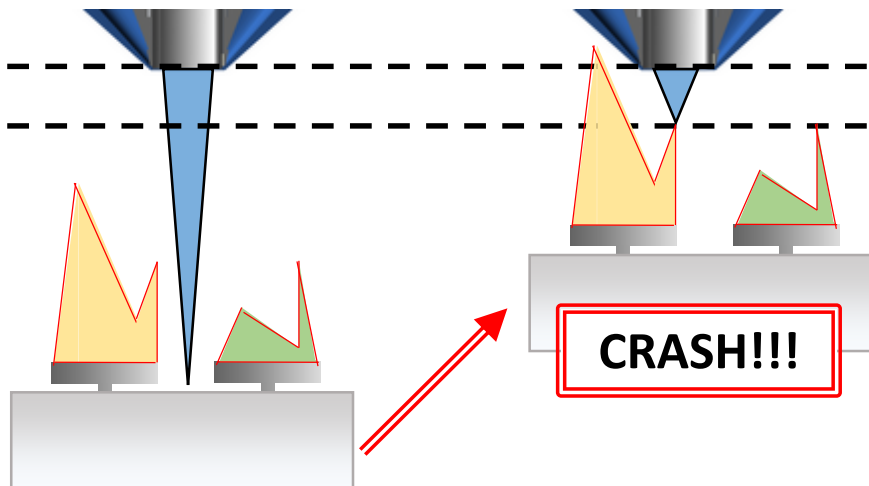
Wrong Sample + Wrong Position



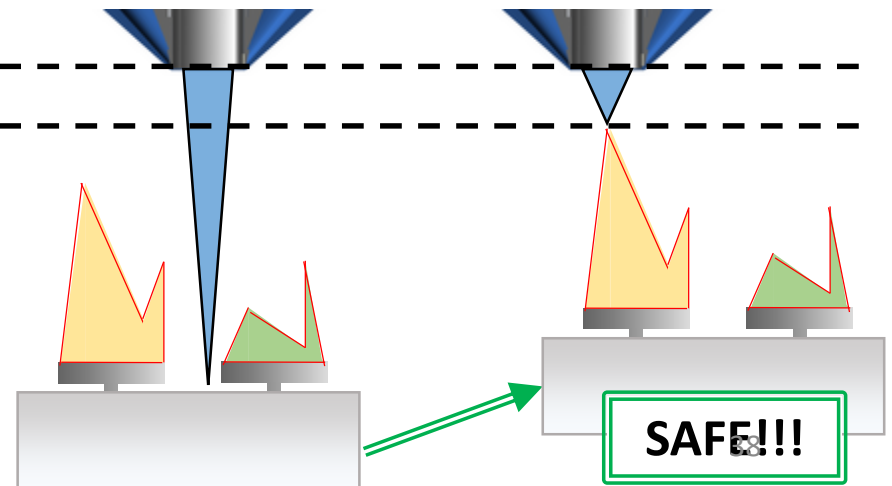
Wrong Sample + Correct Position



Correct Sample + Wrong Position



Correct Sample + Correct Position



XI. Working Distance – 3/3

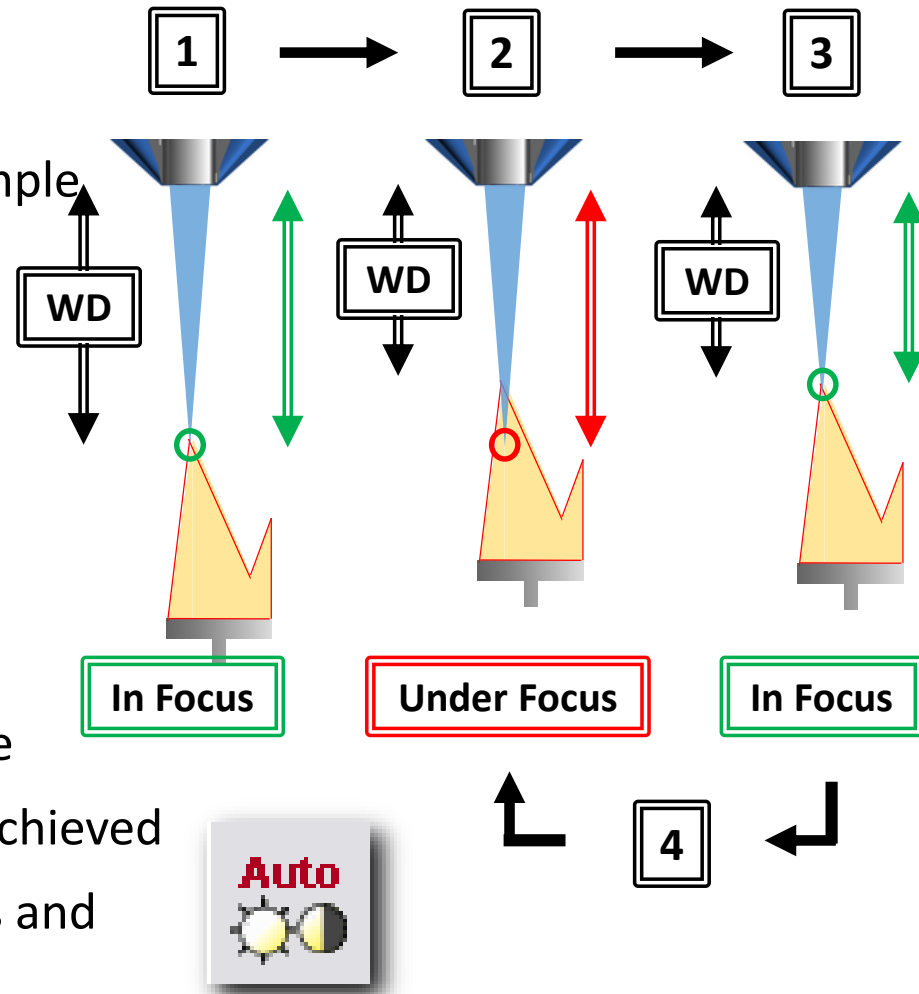
PROCEED WITH CAUTION AS CHANGING THE WORKING DISTANCE CAN RESULT IN DAMAGE TO THE SEM!

1. Identify current WD by focusing on sample
2. Raise the specimen stage by SLOWLY turning the Z-knob counter-clockwise



3. Identify new WD by focusing on sample
4. Repeat steps 2 - 3 until desired WD is achieved
5. Click **Auto** to auto adjust the brightness and contrast if too dark when necessary

6. SLOW DOWN WHEN YOU REACH ~ 10 mm AND DO NOT GET LESS THAN 5 mm



XII. Image Preparation – 1/2

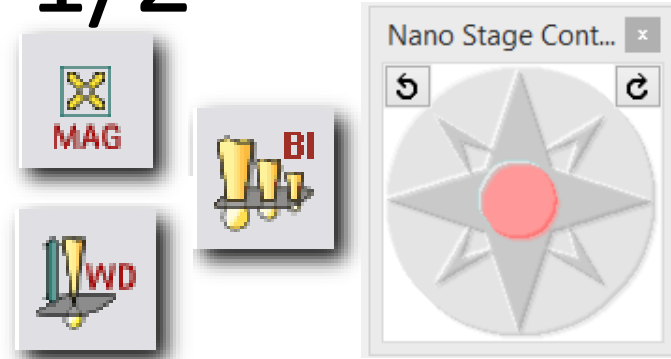
Imaging at **MAG** \geq 10 kX requires optimization steps **XIII. Column Centering** and **XIV. Stigmation**

Correction after completion of **XII. Image**

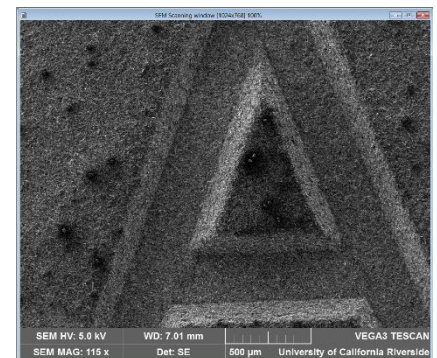
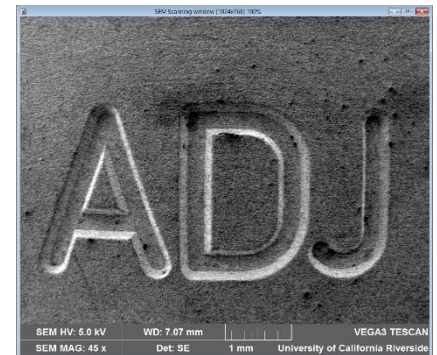
Preparation, else skip and proceed next to **XV. Image**

Acquisition directly

1. Right-click on **MAG** and select **Minimum Magnification** to see your whole sample again
2. Identify an area of interest on your sample to image by using a combination of **MAG**, **Stage Control**, focusing (**WD**), and **BI**



Example



XII. Image Preparation – 2/2

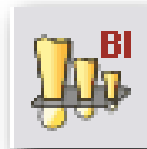
- Bring the area of interest to the **center** of SEM scanning window and to the **highest desired magnification** (e.g. Desired Mag = 10 kX)

You will **NOT** use the **Stage Control** after this step, so **ENSURE** that the image at the Desired Mag is the one you wish to take before continuing

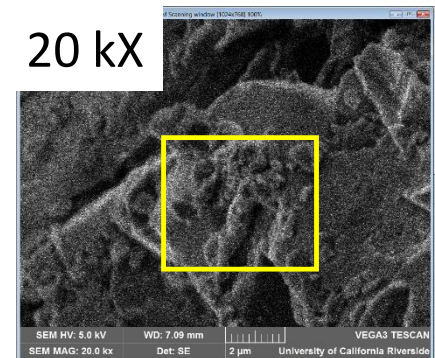
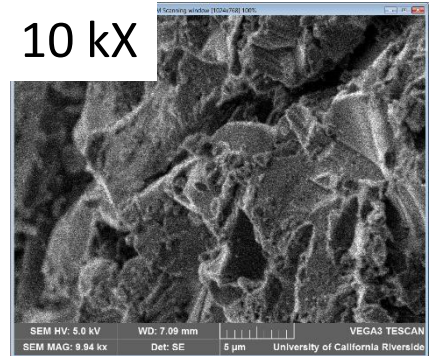
- Increase **MAG** by $\geq 2X$ the desired Mag using the Pad (e.g. New Mag = 20 kX, 30 kX, etc...)

Higher **MAG** yields better results but gets more difficult to optimize

- Reduce **BI** if necessary to increase resolution
- Change scan **SPEED** to 3 or 4 to remove graininess
- Focus (**WD**) your sample again



Example

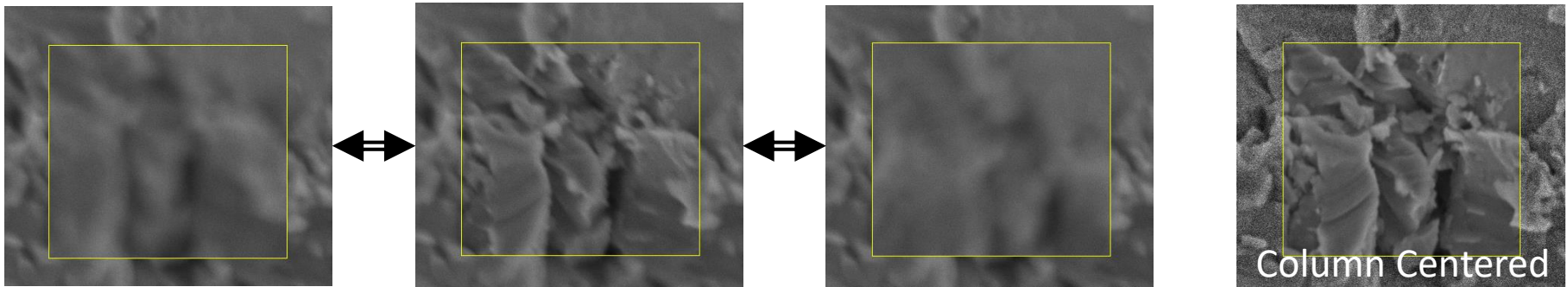


Recommended Initial **BI** values

Magnification	Beam Intensity
Min → 200	18 → 13
200 → 2000	12 → 8
2000 → 10k	10 → 7
10k → Max	7 → 4

XIII. Column Centering – 1/3

1. Create a **Focus Window** around a feature of interest
2. Click **WD** and bring the feature into focus
3. If image moves or shifts as you focus, then column centering needs to be completed and continue to **Step 5**
4. If image does not move or shift, proceed to **XIV. Stigmatism Correction**



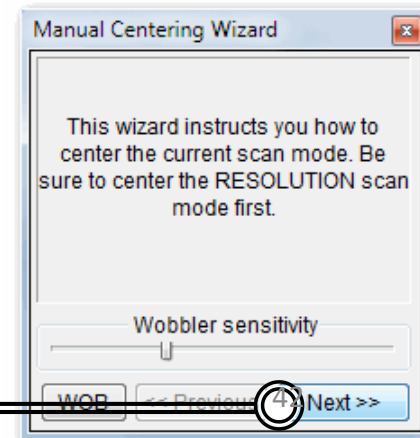
Under-focused

Focused

Over-focused

Column Centered

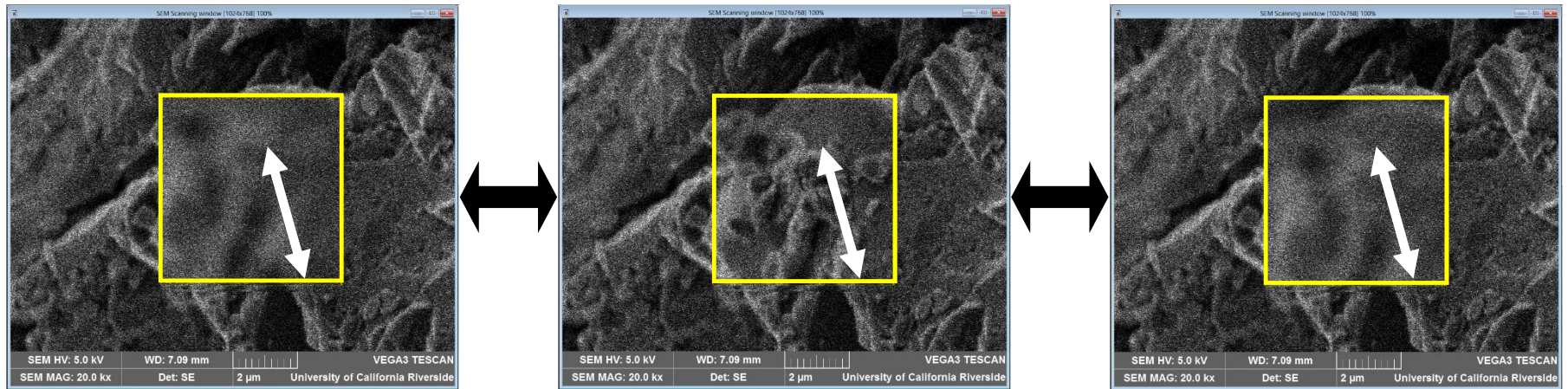
5. Click **Manual Column Centering** button
6. The Manual Centering Wizard will appear
7. Click **Next>>**



XIII. Column Centering – 2/3


8. Your image will now “wobble” in and out of focus

If image has any X or Y translation as it wobbles, you will need to remove it



9. Minimize image movement by adjusting the OBJ Centering using the trackball

X: Hold F12 +  trackball

Y: Hold F11 +  trackball

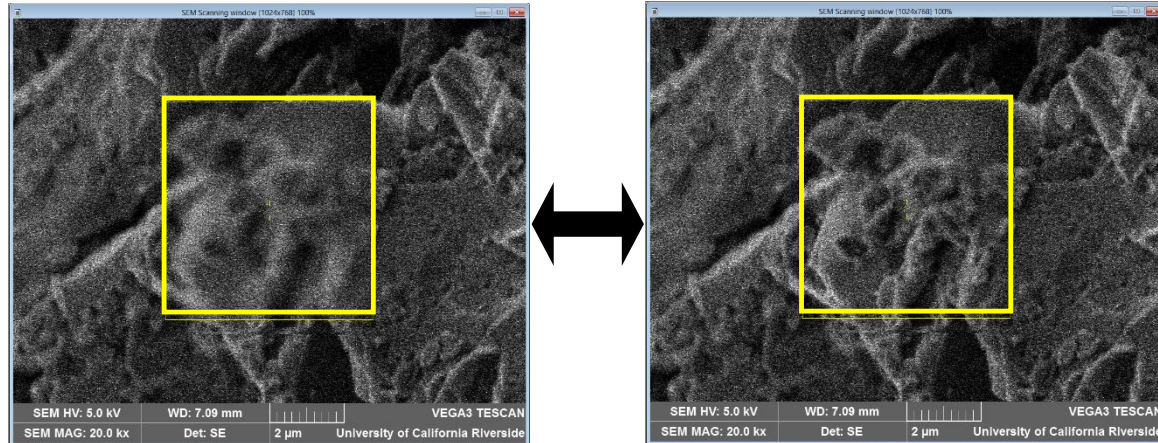


= Change only X-movement

= Change only Y-movement

XIII. Column Centering – 3/3

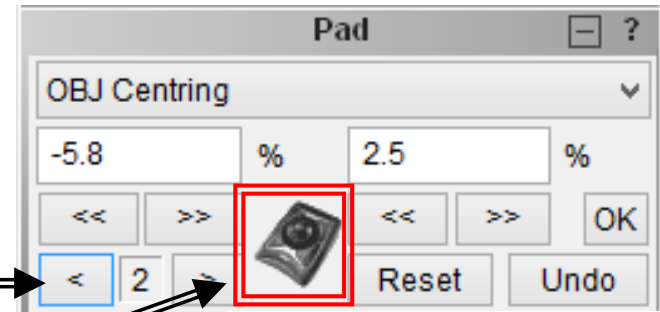
8. The image should remain stationary with no X or Y translation but only oscillate in/out of focus



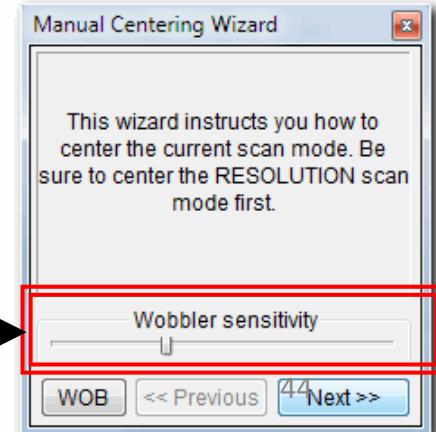
9. Adjust the sensitivity to finely control the **OBJ Centering** if necessary

Recommended Value = 5 first then 2

10. If flashing trackpad is present, click << **Previous** and **Next** >> to reset



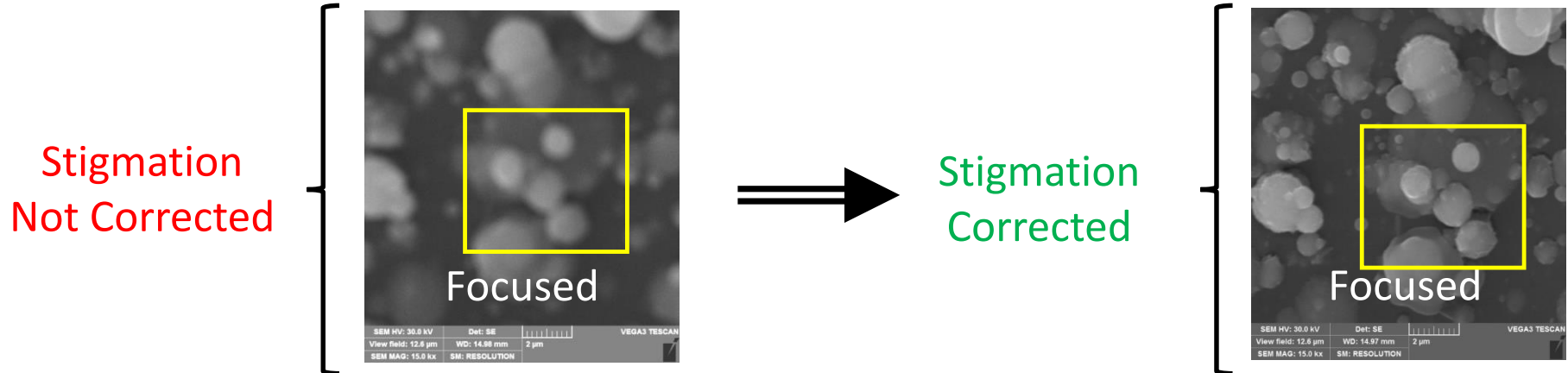
11. Adjust the **Wobbler sensitivity** to change the extent of “wobble” if necessary at very high magnifications



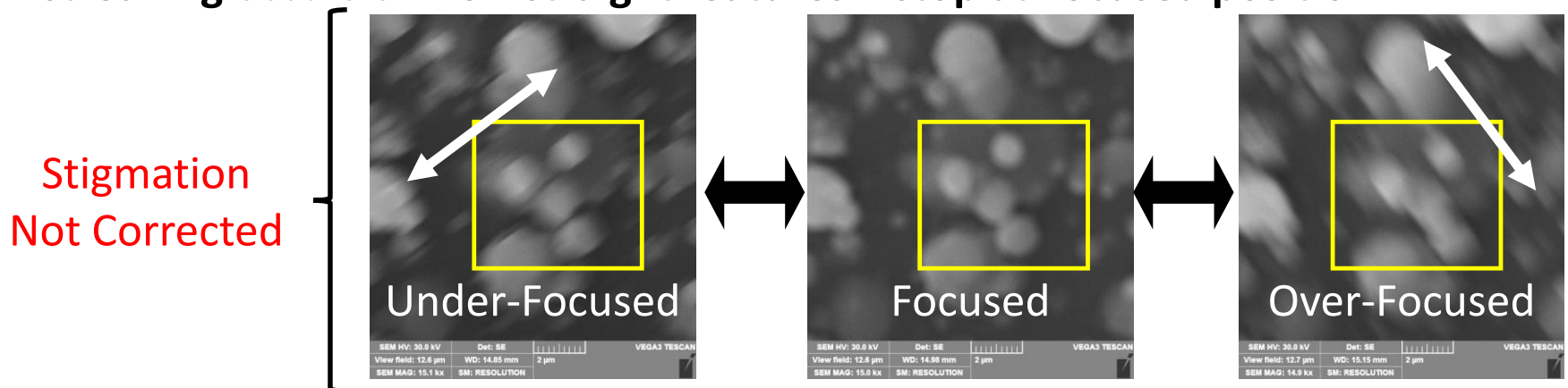
12. Click **Finish** when done

XIV. Stigmation Correction – 1/4

1. Create a **Focus Window** on a feature of interest
2. If Stigmation corrected, a focused image will become **significantly sharper**



3. Click **WD** and bring the feature **in and out-of-focus** (both sides) to check if any **streaking** occurs on **non-straight features** – stop at **Focused position**



4. Any streaks are evidence that **Stigmation Correction** is necessary

XIV. Stigmatism Correction – 2/4

5. Set **SPEED = 4** + appropriate **BI** (see table)

6. Click **WD** and create a **Focus Window**

7. Focus on a feature (**WD Sensitivity = 2**)
as **BEST AS YOU CAN**




8. Click the **STG** to set as active function




9. Set **STG Sensitivity = 6** (slow down trackball for accuracy near “sweet spot”)

10. Achieve a sharper image by adjusting the Stigmators one at a time (X and Y)

X: Hold F12 +  trackball



= Change the X-component

Y: Hold F11 +  trackball

= Change only Y-component

11. **CAREFULLY AND SLOWLY** adjust each Stigmator component (X and Y) until you can identify the “*perfect*” or setting with the sharpest image

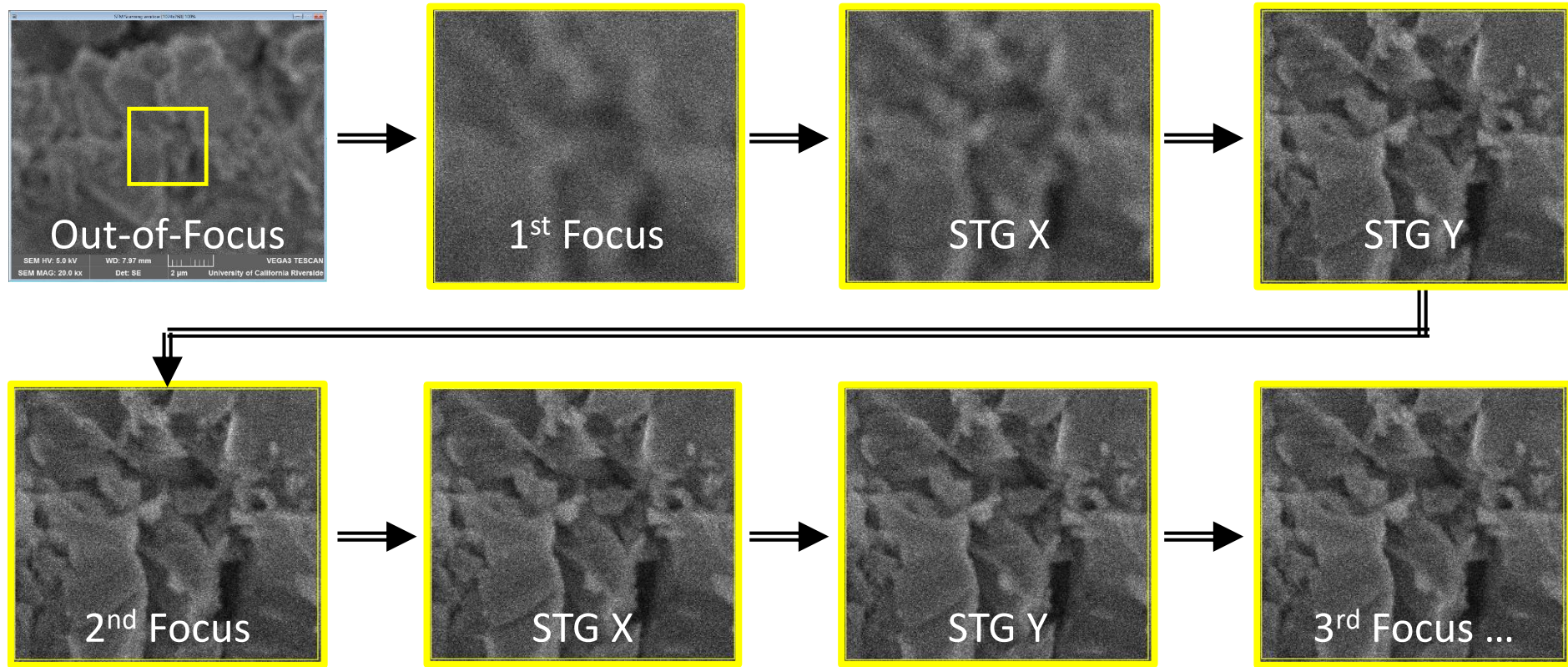
12. **REPEAT Steps 6 – 11** until you no longer see any improvement in sharpness

Recommended Initial **BI** values

Magnification	Beam Intensity
Min → 200	18 → 13
200 → 2000	12 → 8
2000 → 10k	10 → 7
10k → Max	7 → 4

XIV. Stigmation Correction – 3/4

13. If your image still doesn't look "good", 99% it's because of poor **STG Correction**
14. The sequence of **STG Correction** should resemble the following:

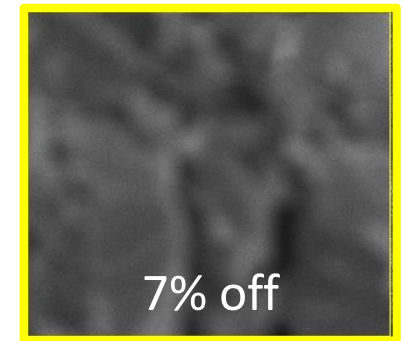
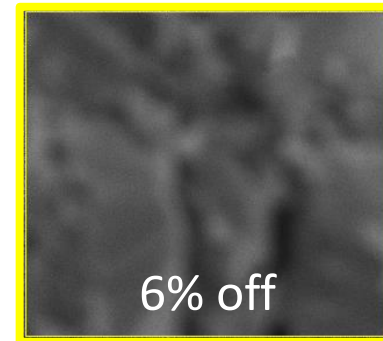
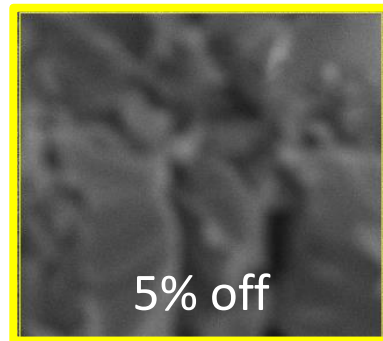
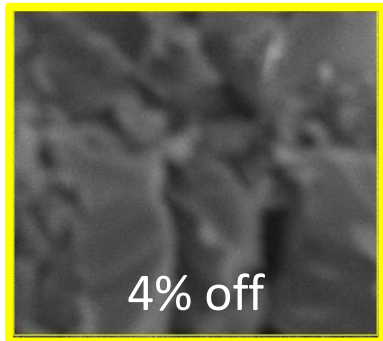
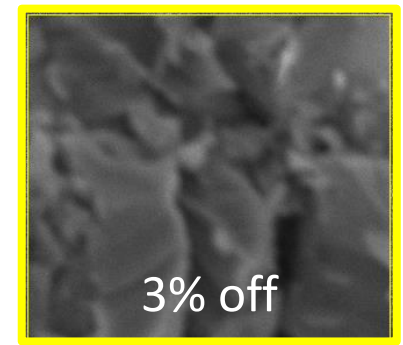
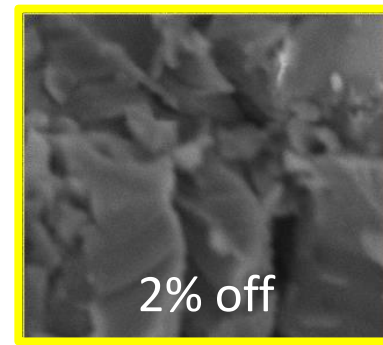
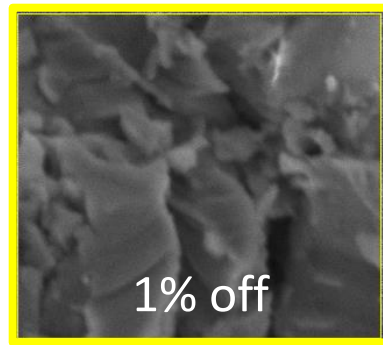


15. Repeat the sequence as necessary until the image looks "good"

XIV. Stigmatism Correction – 3/4

16. Proper *STG Correction* is **EXTREMELY** sensitive

17. A few % values off from “perfect” setting, and your image will look very blurry!



18. If this is the case, **GO BACK AND RE-DO** the *STG Correction*!

XV. Image Acquisition – 1/3

1. Create **Focus Window** and achieve the **BEST** focus (Recommend Sensitivity = 2)



(Do **NOT** focus again **AFTER** this step!)

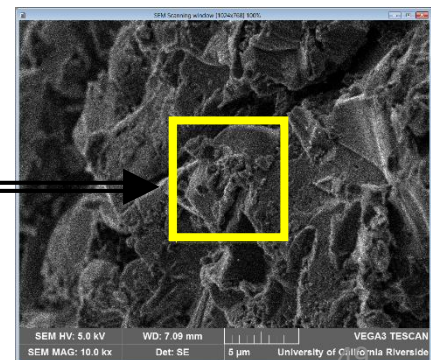
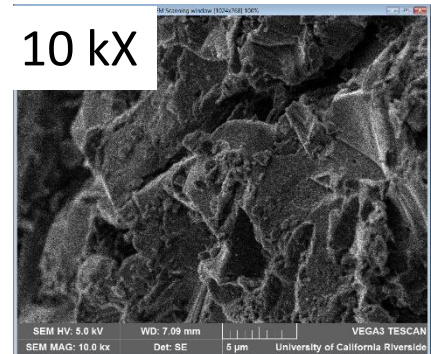
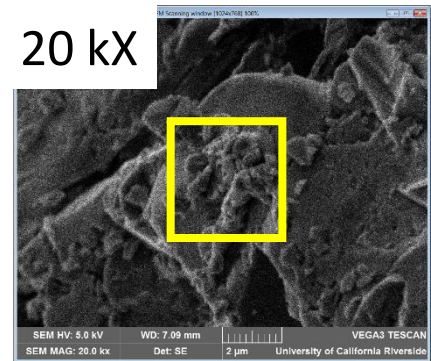
2. Click **MAG** and set back to desired magnification (e.g. Desired Mag = 10 kX)



3. Activate the **Focus Window** over a desired feature

Smaller window = requires less time to refresh

Example

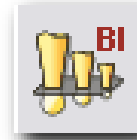


XV. Image Acquisition – 2/3

4. Identify maximum **Acquisition Time** for your image (e.g. 2 min) and select corresponding **Speed** (e.g. **SPEED** = 7)



5. Adjust the **BI** until a balance between resolution is matched with graininess



6. Click **Auto** to auto adjust the brightness and contrast as you change the **BI**



SPEED	Acquisition Time
1	0.12 sec
2	0.30 sec
3	0.87 sec
4	3 sec
5	16 sec
6	32 sec
7	1 min 36 sec
8	4 min 34 sec
9	13 min 58 sec
10	44 min 4 sec

NOTE: Remove focus window first else it will only adjust pixels found within focus window + **change speed back to 1** for faster auto correction

7. If high resolution is desired but excessive graininess is present, increase the **Acquisition Time** (e.g. **SPEED** = 7 -> 8)
8. Repeat Steps 5 – 6 until desired balance between resolution and graininess and is achieved (e.g. see next slide for examples)

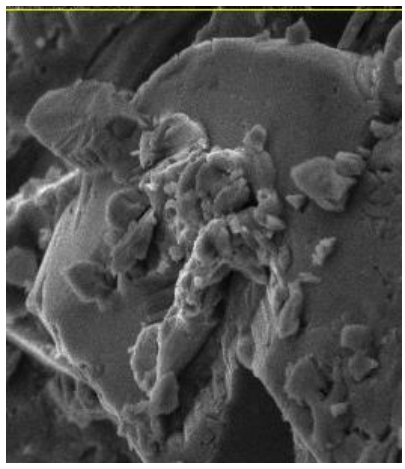
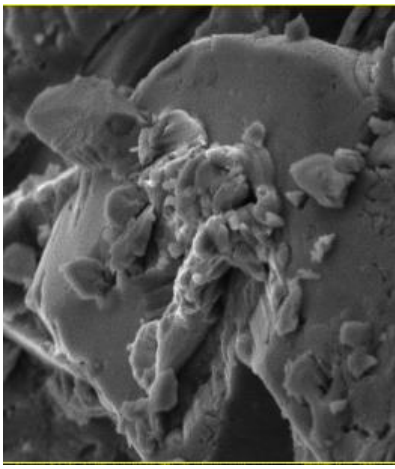
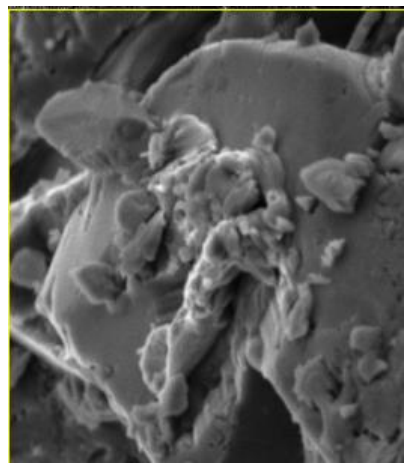
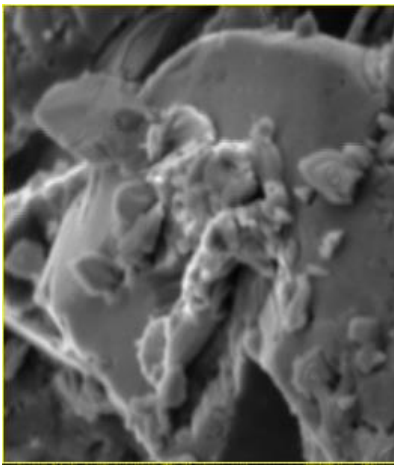
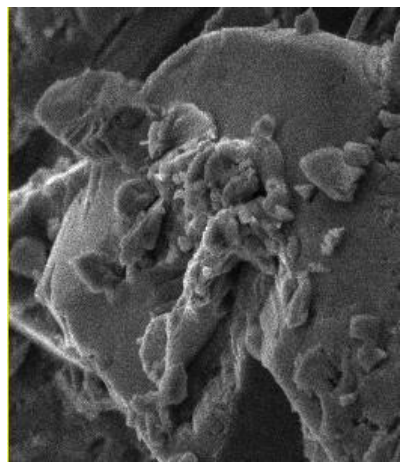
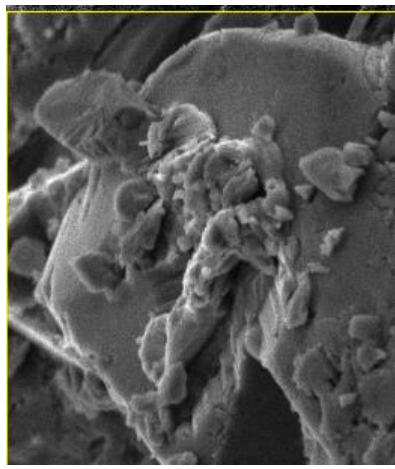
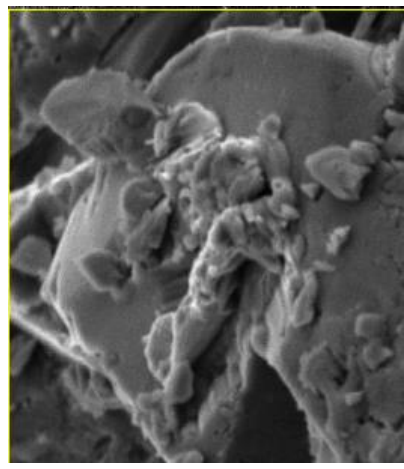
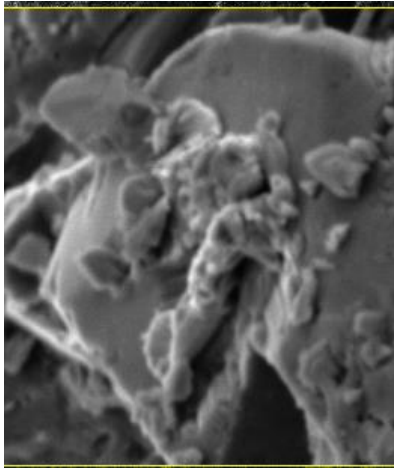
XV. Image Acquisition – 3/3

Low Resolution
Low Graininess

High Resolution
High Graininess

==== 10 ===== 8 ===== **BI** ===== 6 ===== 4 =====>

==== 7 =====
Speed
==== 8 =====>



XVI. Saving – 1/1



1. Click **Acquire** to capture image
2. If desired, you may save information to the image file

Note = the basic description

Sign = the enlarged description

Description = the detailed information

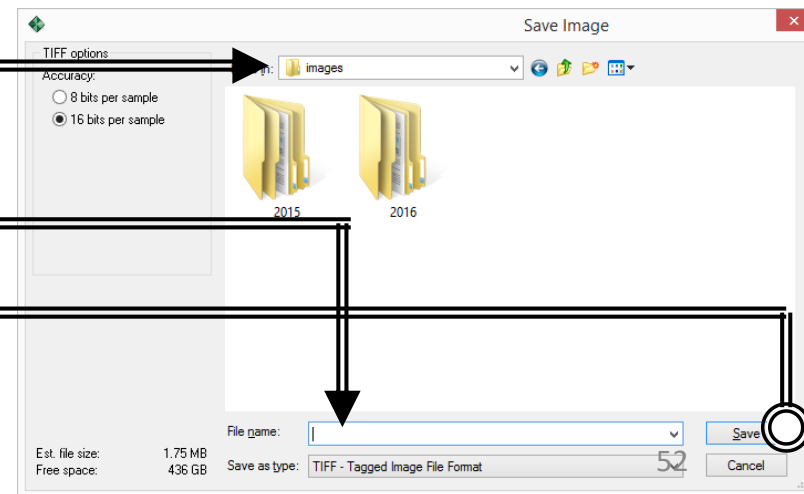
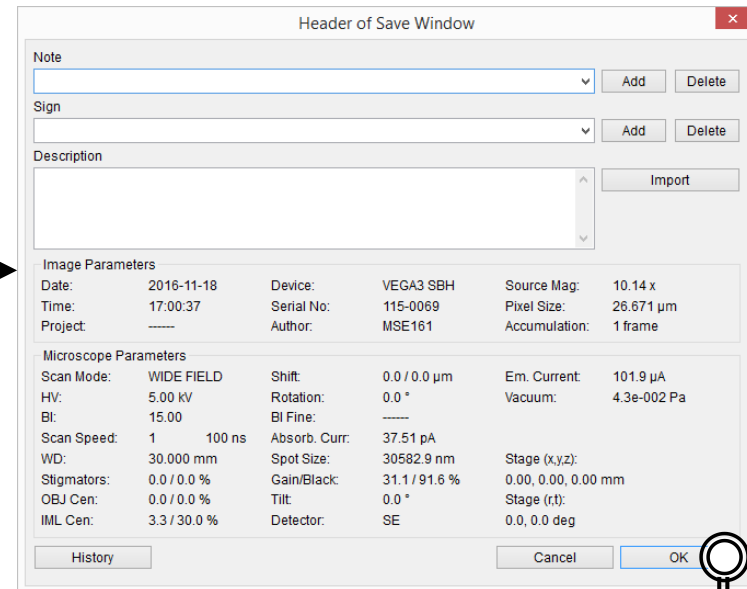
Add = saves the Note or Sign in the list

Delete = deletes the Note or Sign from the list

3. If you choose not to include any Header information, click **OK**
4. Choose the folder location for your images

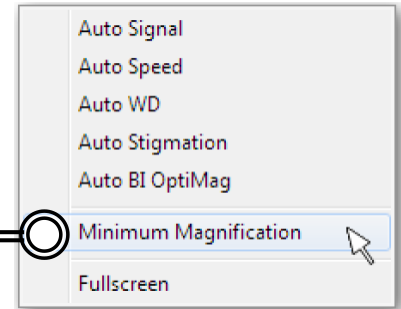
5. Enter your file name

6. Click **Save**



XVII. Sample Unloading – 1/3

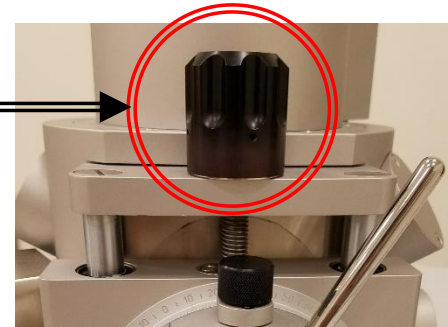
1. Right-click on **MAG** and select **Minimum Magnification**



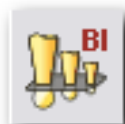
2. Click **SPEED** and select **SPEED 1**



3. Carefully **Lower** the sample stage to the lowest position by turning the **Z-knob clockwise**



4. Set **BI** to 15

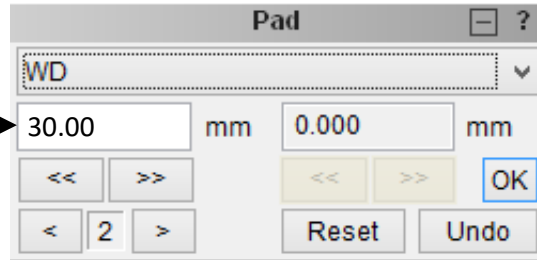


5. Click **Auto**

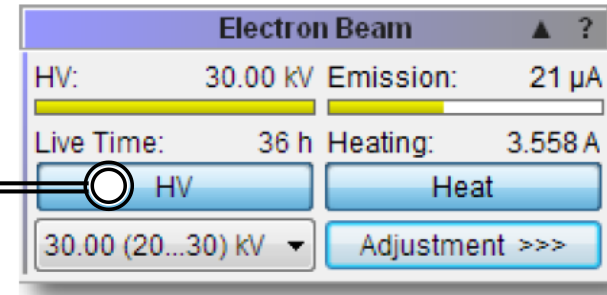


XVII. Sample Unloading – 2/3

6. Set **WD** to 30 mm

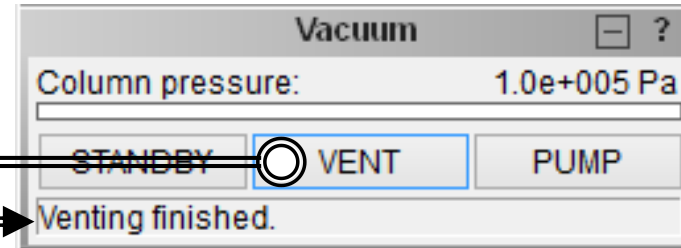
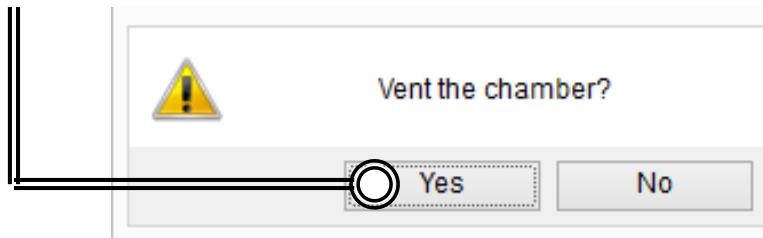


7. Click **HV** to turn off the high voltage



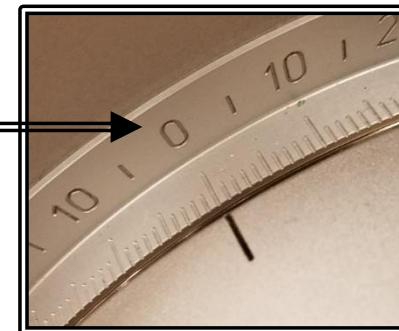
8. Click **VENT** to vent the microscope

9. Click **Yes** to confirm venting



10. Wait until "**Venting finished**" appears

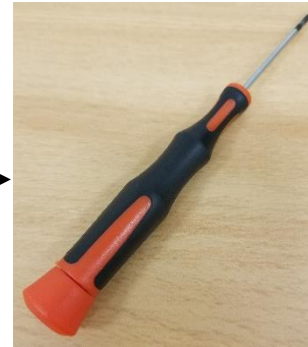
11. Set the tilt of the specimen stage back to 0° if not already set to 0°
(Advanced Users only)



XVII. Sample Unloading – 3/3

12. Gently pull the chamber corners toward you to open the chamber

13. Loosen the screw holding the specimen stub on the specimen stage



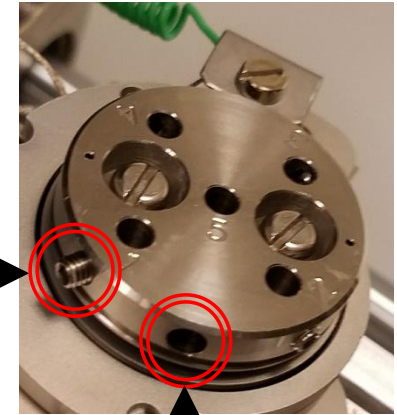
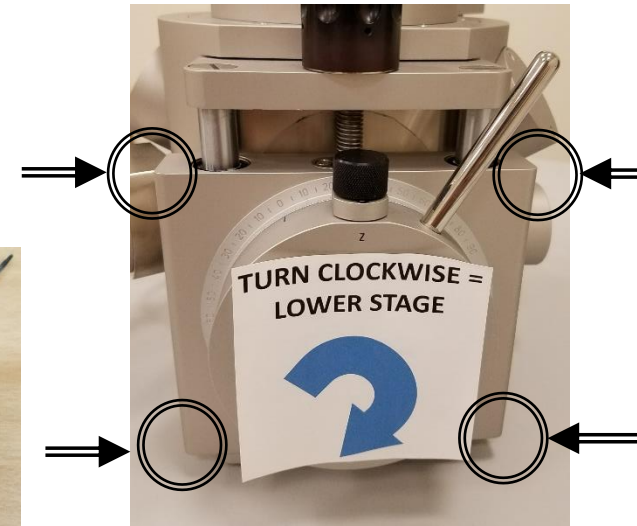
14. Rotate the stage if necessary to access screw port



15. Using the provided tweezers, carefully remove the specimen stub out of the specimen stage



16. Tighten the screw back so it doesn't stick out



XVIII. Cleanup – 1/1

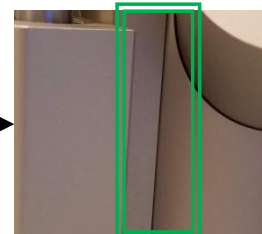
1. Ensure sample stage is at lowest position



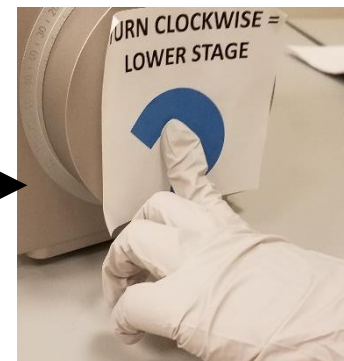
2. Check O-ring and ensure that it is sitting inside the recessed groove



No Gap



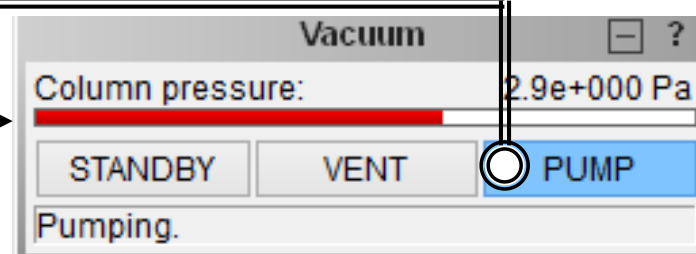
3. Carefully close the chamber door and check there is no gap



4. Place finger against chamber door

5. Click **PUMP** to start pumping down chamber

6. Wait until bar graph shows **red** to release finger



7. Wait until the bar graph turns **green** or "**Vacuum ready**" appears (~ 3 min)

8. Open **File** menu and select **Logoff**, click **Yes**

9. **Record** your total time used in **Sign-in sheet**

