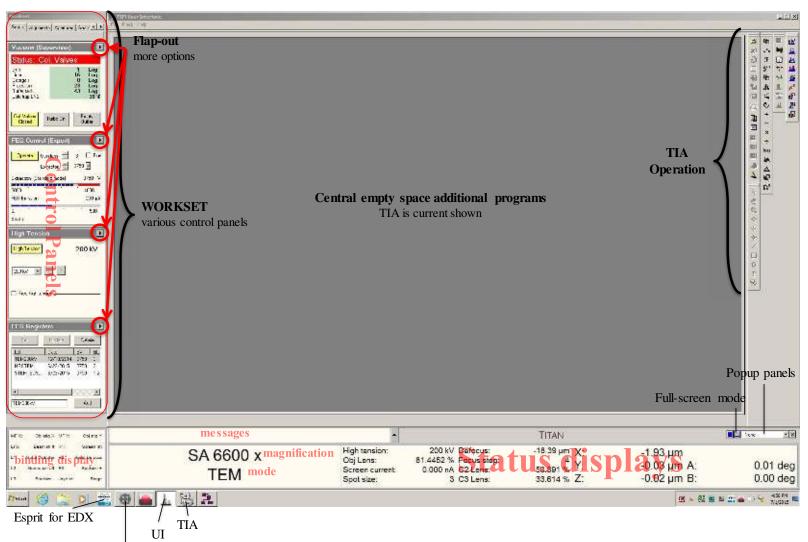


# 200 kV Titan Themis (S)TEM with Cs Image Corrector

By Tong Wang, Sheng Zhang CUNY ASRC Imaging Facility



## **User Interface Overview**



Cs corrector program

TITAN Themis is 200 kV high resolution TEM, whose interface contains three parts: TEM server, User Interface (UI), and Control pads (left & right). Users interact with TEM server via UI. An overview of UI is show above. UI contains elements that can be divided into three categories:

- Main program: program title and menu bar.
- Control panels: control panels are grouped in worksets that can be selected via a tab at the top of the area with control panels.
- Information panels: binding, messages, status displays.

The central empty space is reserved for other programs (e.g. TIA, Esprit).

Notes: Position the mouse cursor over a particular panel and press F1 on keyboard to invoke the UI help pages for that particular panel.



#### **Microscope Control Pads Overview**

Control pads (left & right) consist of a set of standard TEM controls:

- Left: Tilt, user beam shift trackball, Intensity (Brightness), Stigmator, Multifunction X (MF X), programmable buttons (L1, L2, L3).
- Right: Diffraction, Wobbler, Eucentric Focus, Multifunction Y (MF Y), Magnification, Focus, Focus Step, Joystick, Z height, programmable buttons (R1, R2, R3).

MF X/Y knobs' default function is "user beam shift X/Y" and can be assigned (bind) to other functions. Programmable buttons are user-defined buttons and can be assigned (bind) to frequently used functions.



# **Preparation & Start-up**

 Log in using supervisor account (Optional). The microscope server control software (Microscope Software Launcher) should automatically starts. Click the start icon to start microscope UI. Launch other programs (e.g. TIA)



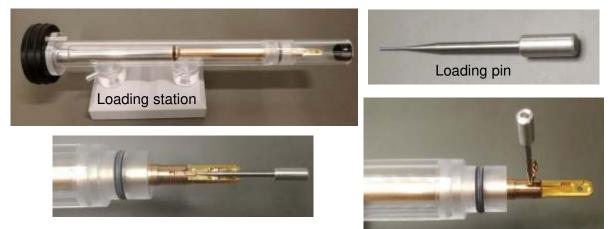
microscope UI. Launch other programs (e.g. TIA, Cs image corrector) if needed. UI software launching progress can be monitored by click **window>message**.

- 2. Check the liquid nitrogen level (LN2) in cold trap (**Setup>Vacuum**). Please notify Imaging Facility staff if it is below 20%.
- 3. Make sure that column valves are closed: **Col Valves Closed** button in **Setup>Vacuum** is in yellow color.
- 4. Make sure that camera is retracted: **Insert** button in **Camera**>**CCD/TV Camera** is in grey color.
- 5. Make sure the stage is reset: values for X, Y, Z, A, and B in TEM status window at the lower right corner of UI are zero or close to zero. If not, click **Holder** in **reset** panel of **Search> Stage>Control** page to reset **Holder**.
- 6. Make sure the objective lens aperture is retracted. Objective lens aperture control can be found in **Apertures**.



## Load Sample

1. Load TEM grid to single or double tilt holder using holder station. Note: Put a washer on top of the grid when using the double-tilt holder. If doing EDX, put the sample side facing down (e.g., the side with sample applied on) using the double tilt holder. After loading, double check the sample and washer won't fall off.



- 2. For samples prepared by chemical solvent, it's suggested to use plasma etcher in the Imaging Suite to clean the TEM grid within the holder together (70% power, 20 to 30 s).
- 3. Log in and enable the microscope in Badger system.
- 4. Make sure Turbo pump is off ("Turbo On" is grey). Insert holder into stage half way in by aligning <u>guide</u> <u>pin</u> on holder (see picture below) at 5 o'clock position on the stage ("Close") until you feel some resistance. Push the holder further in for 8 mm with a slight force, sometimes need to twist the holder a little to find the right angle to push further inside. The holder should be firmly inside the hole. This will trigger turbo pump to pump the airlock (turbo pump button will turn orange and then yellow, 3 mins counting down starts). If using double-tilt holder, connect the cable to the CompuStage.



- 5. Select holder in the message tab of UI (single tilt, double tilt, etc.) and confirm.
- 6. After 3 min counting down, turn holder <u>counter-</u><u>clockwise 150°</u> and guide holder fully into the column. Note: after turning 150°, apply an outward force to guide the holder into the column, DO NOT let the vacuum pulls the holder in freely.
- 7. Check the vacuum of Octagon and wait until the value is less than 20 log. Note: the vacuum of Octagon may show value below 10 log after inserting sample holder, you still need to wait at least 3 mins before opening column values.





### **TEM Alignment**

1. Click **Col Valves Closed** button to open column valves, **Column Valves Closed** button should change from yellow to grey and you should observe beam on the fluorescence screen. In case you cannot find the beam, reduce the Magnification and move the stage in X-Y plane using Joystick.

Find a visible feature on the grids (e.g., a cluster of particles) and center it in the view. Check the "Coarse" above **Intensity** on the left control pad is activated.

2. Adjust Eucentric Height by one of the following methods:

Vacuum (Supervisor) . Status: Col. Valves Gun Log 1 Liner 16 Loa Octagon Log 5 Projection 21 Log Log Buffer tank 42 Cold trap LN2 69 % Col. Valves Empty Turbo On Closed Buffer

Method 1: Center a recognizable feature on the sample, activate **Alpha Wobbler** in **Search>Stage>Control** (see picture below), changing Z height to minimize the movement of sample image, increase the magnification and repeat Z adjustment until above SA 20000 x. Then deactivate **Alpha Wobbler**.

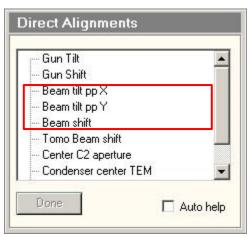
Stage <sup>2</sup>	Control File St
Go Add Update Delete	Positions / tracks     Delete All   Clear Tracks     Stage control     Power step (1/8 8)   1     XY separately above   500000   x
SX1 SX2   I I <td< th=""><th>Reset     Holder   XY   AB     Alpha wobbler   Wobbler     0   5   10   15    </th></td<>	Reset     Holder   XY   AB     Alpha wobbler   Wobbler     0   5   10   15



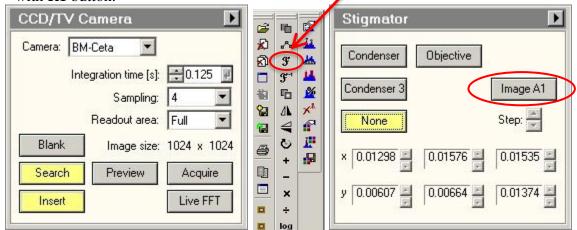
Method 2: Center carbon film region in the view. Press **Eucentric Focus** on right control pad once, change Intensity to make the beam a minimum spot, then adjust Z height to minimize halo around the beam spot at a magnification above SA 20 kx.

Method 3: Press **Eucentric Focus** once, expand beam to show sample image, adjusting Z height to minimize image contrast.

- 3. Direct Alignments in "Apertures" Tab:
  - a. Change magnification to ~ 310 kx in SA range.
  - b. Press **Eucentric focus** button on right control pad.
  - c. Condense the beam to a spot using Intensity. Activate Beam tilt pp X in Aperture>Direct Alignments, bring the two spots together using MF X/Y knobs. Repeat the same procedure to tune Beam tilt pp Y. (If beam is out of the view, change to lower magnification, activate Beam Shift and use MF X/Y to move the beam back to the center.)



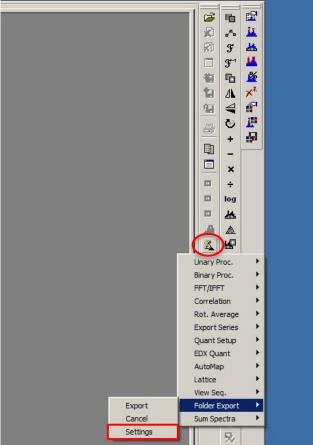
- d. Activate **Beam Shift** and use **MF X/Y** to center the beam. (Do not use the trackball on left control pad to move the beam.)
- e. Click "Done" to finish **Direct Alignments**. Spread beam **Intensity** to cover full fluorescence screen.
- 4. In **Camera**>**CCD/TV Camera**, click **Search** and **FFT** buttons. Lift the fluoresce screen with **R1** button.



(Optional) Find a pure carbon area (amorphous carbon support film) without any sample of interest. If the FFT rings are not circular, activate **Image A1** in **Stigmator** (**Apertures** tab) of UI and use **MF X/Y** to manually adjust A1 stigmatism by making the rings circular (change focus to make the rings bigger). Click **None** after done. Perform Cs Image Correction if you plan to image HR-TEM at atomic resolution (instruction on the following pages).



- 5. Find areas of interest, adjust Focus to bring the sample close to focus (no rings on FFT), press R2 button to reset focus value. Adjust defocus and take images by clicking Acquire in Camera>CCD/TV Camera. Click Save as or Auto Save to save the images in \*.emi format. Alternatively, right click on the image and select Export, save the image as tif file with scale marker and full resolution.
- 6. Folder Export all \*.emi files: after capturing all the images on the sample, locate the Component icon in the TIA. Select Settings in Folder Export, change both Source folder and Target folder to where you saved \*.emi files (Y:\Users\.....). Then click Export in Component > Folder Export to export all the emi files to tif files.

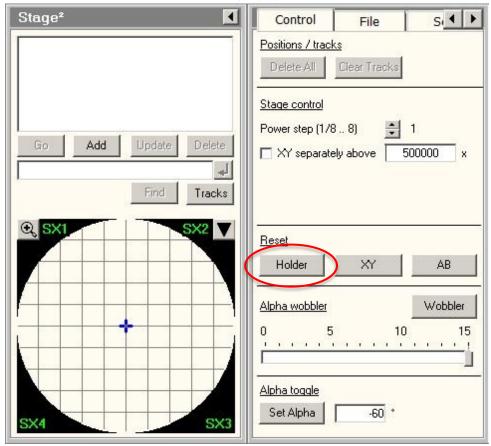


Options	<u>×</u>
Dptions:	
Source folder	Y:\Users
Target folder	Y:\Users
Recurse folders	
Export images	
Image format	PC TIFF w/scale marker (full res)
Store actual resolution in tiff tags	
Export spectra	
Spectrum format	One-column text



### **Shutdown Procedure**

- 1. Return to TEM mode in FEG registers if you performed STEM application.
- 2. Stop **Search** in Camera. Retract CCD (BM-ceta) camera and make sure that the **Insert** button is in grey color.
- 3. Lower the fluorescence screen (**R1** button). Put the rubber cover on.
- 4. Click **Col Valves Closed** button in **Setup>Vacuum** to close the column valves and make sure the button is in yellow color.
- 5. Center the stage: click **Holder** in **Reset** panel of **Search> Stage>Control** page (see picture below) to move stage to center, watch the TEM status window at the lower right corner of UI and make sure that values for X, Y, Z, A and B are zero or close to zero.

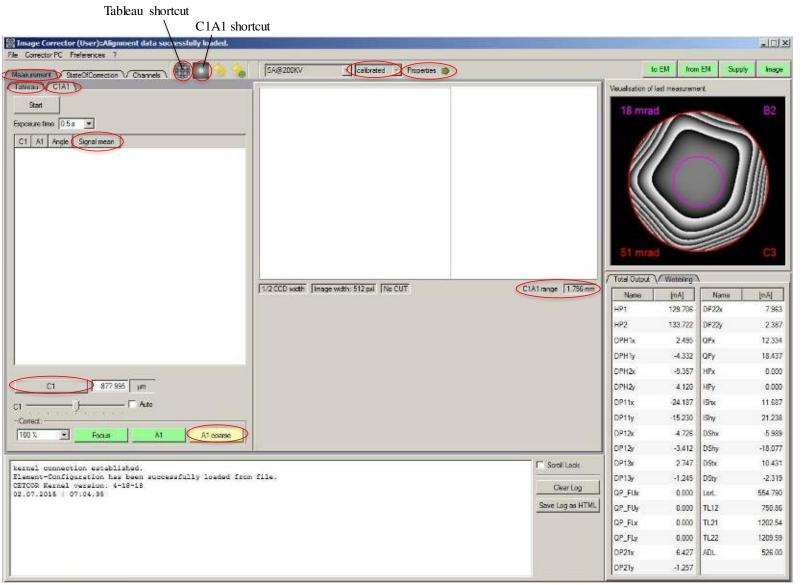


- 6. Retract objective lens aperture and selected area aperture if you ever used one.
- 7. Leave the magnification in the lower SA range (4kx 10kx). This is essential to maintain stable objective lens current and prevent thermal drift for the next user
- 8. Now you can remove the holder out from microscope and unload your grids: Pull out the holder slowly until it stops, turn the holder <u>clockwise</u> until it stops, and pull it out with a little force carefully. Note: Use two hands to pull the holder out of the station: use two fingers of the left hand to gently push against the station (purple region) and use the right hand to pull out the holder, in order to prevent any damage to the station.
- 9. If the Turbo pump starts, turn it off. The "Turbo On" should be grey.
- 10. Repeat from the sample loading process if you have more samples to characterize.
- 11. When you finish, clean up working area. Log out the facility in Badger system.



## **Tuning Cs Image Corrector**

 Click Search and FFT buttons in Camera>CCD/TV Cemera. Find a pure carbon area (amorphous carbon support film) without any sample of interest. Adjust magnification to 310 kx in SA range. This makes sure that the magnification is shown "calibrated" in Image Corrector Tuning program and Nyquist Frequency value (shown in Properties) is about 4~5 (This range of magnification is optimum for tuning corrector). Do Direct Alignments following instructions above. Adjust Image A1 in Stigmator tab of UI to manually adjust A1 stigmatism by making the rings circular (do not need to be perfect at this stage). Stop the camera Search. Make sure the Sampling of Search is 4.



In Image Corrector software, press Start in C1A1 tab, adjust Intensity to make the Signal mean in the range of 6000 ~ 10000. Adjust Focus until C1 value is in the range of -600 nm to -800 nm (underfocus). Alternatively, you can directly apply a negative value (~ 30 % of C1A1 Range) to C1: at the bottom of the measurement window, choose a value for C1 and press C1 button.



- 3. Press A1 coarse to reduce A1 value (obj stig) to be less than 5 nm.
- 4. Go to Tableau tab, select Tableau type to "Fast", Outer Tableau tilt [mrad] to 16 (or 18), and exposure time to 0.5 s. Note: In Tableau, beam is tilted to collect series of images to do measurements, when beam is tilted TEM is in dark field mode.

essurement V StateOfCorrection V Channels ) 🏥 🌅 🧆 🐁 🛛 SAGE200KV 🔄 Calibrated 🔄 Properties 🐡		to EM fm	m EM Suppl	y3 Int
otkey/CIAT	Vecalizat	on of last measurem	nent.	
Stat Options (a) Fast Exposure time 0.5 s (c) Standard Outer tableau til (mod) 16 Enhanced Angle Confidence First order C1 845m - 2.43m Sgma all	18			
A2 20.1m 146.2' 48m 82 29.4m 101.5' 27.8m C3 -6.3m - 2.35m	51	tput V Webbing		0
33 306mm 45.5" 1.38µm Signa used 1/2 CCD width Image width: 512 pvl No CUT	C1A1 mage 1.756 mm Nam		Name	jm?
33 169rm -126.8° 794rm - C1 A1 Angle Signal mean	HP1	129.706	and the second s	7
4 46.1µm -179.8° 43.4µm Percent	HP2	133,722		
	DPH1x	2.495	A CONTRACTOR	12
	DPH1y	-4.332	QPy	18
	DPH2x	-9.357	HPx	3
			HPy	
	DPH2v	- 4. 12W	1.000	
	DPH2y DP11x	4.120	ISTA	
	DP11x	-24.187		
	DP11x DP11y	-24.187 -15.230	IShy	2
a No Data	DP11x	-24.187	19hy DShx	2
	DP11x DP11y DP12x DP12y	-24.187 -15.230 -4.726	19hy DShx	2 -1
nel connection established. rent-Configuration has been successfully loaded from file.	0P11x 0P11y 0P12x 0P12y	-24.187 -15.230 -4.726 -3.412	IShy DShx DShy DShy	2 -1 1
nel connection established. cent-Configuration has been successfully loaded from file. COR Kernel version: 4-18-18	DP11x DP11y DP12x DP12y DP12y DP13x	-24.187 -15.230 -4.726 -3.412 2.747 -1.245	IShy DShx DShy DShy DShy	2 -11 11
nel connection established. cent-Configuration has been successfully loaded from file. COR Kernel version: 4-18-18	Centilock Centilock Centilock Centilock Centilock Centilock Centiloc	.24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000	19hy DShox DShy DShy DShy LorL	2 -11 -11 -11 -15
nel connection established. cent-Configuration has been successfully loaded from file. COR Kernel version: 4-18-16	DP11x DP11y DP12x DP12y DP13y Clear Log DP13y CP_FU	.24 187 -15.230 -4.726 -3.412 2.747 -1.245 0.000 0.000	IShy DShx DShy DShy DShy LorL TL12	2 -11 -11 -11 -11 -11 -11 -11 -11 -11 -1
nel connection established. cent-Configuration has been successfully loaded from file. COR Kernel version: 4-18-16	Clear Log Serve Log as HTML	.24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000 0.000 0.000	IShy DShx DShy DSh DSh DShy LorL TL12 TL21	2 -11 -11 55- 7 121
a No Data Accept mel connection established. ment "Configuration haw been successfully loaded from file. (COR Karnel version: 4-18-18 07.2015   07:04:35	Ceer Log Serve Log as HTML OP 11y DP 12y DP 12y DP 12y DP 12y DP 12y DP 12y DP 12y DP 12y DP 13y DP 12y DP 13y DP 13y DP 13y DP 13y DP 12y DP 13y DP 12y DP 12y DP 13y DP 12y DP 13y DP 13y DP 12y DP 13y DP 12y DP 13y DP	.24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000 0.000 0.000	IShy DShx DShy DShy LorL TL12 TL21	1" 2 -18 -18 -18 -18 -18 -18 -12 -12 -12 -5 -5 -5 -5 -5 -5 -5 -5 -5 -5 -5 -5 -5

- 5. Press **Start** to measure B2 and A2. If A2 is larger than 100 nm, B2 is larger than 50 nm, or either value is larger than Confidence, press **Accept**. Otherwise go to step 7. Note: the parameters highlighted in yellow (except C1) are above Confidence and can be further tuned. Others not highlighted are below Confidence and do not need further tuning.
- 6. The program will automatically enter correction page. Select correction power (100%), press B2 or A2coarse (not A2) whichever needs to be tuned based on the results of step 6.



Then press shortcut to start Tableau measurement again, repeat Step 5 to 6 until the parameters are less than or close to confidence values.

		(User)::Alignme	ent data succ	essfully load	ed.													
_	rector PC Pre	were service		$\mathbf{\mathbf{b}}$	h ch	SA@20	nkv/	- calbrated	- Propert		0				10	to EM	n EM Su	pply ime
55.1	ement \/ Sia	Consider a Constant of Constan	Channels \	$\cup$			_				Des				Vaualisation of			
	Value		Changes	Rel.sze		Value	e balqecos te Angle	Confidence	- 44	- 32-	Value	vicus measu Angle		Confidence				
1	2.043 µm	nive	880	242 %	C1	-843.5 rm	7.90	Opm	290 min	c1	Table		2	Carriellive	18 mra	d		B
	15.95 nm	-29.5 *	3508	1864 %	AI	855.4 pm	-39.5 *		290 min	AL					1		-	
					-	and the second		0 pm										
	16.09 nm	146.7 '	0	100 %	A2	16.89 nm	146.7*	42.97 nm	290 min	A2								1 114
	26.7 ntt	-138.2 *	2190	69 %	82	38.82 nm	-144.9 *	24.87 nm	290 min	82								111
	6.424 µm	1	0	100 %	63	-6 424 µm	(tr)	2.105 µm	290 min	C3							1	
	550.3 nm	60.3 °	1748	100 %	A3	560.3 rm	60.3 °	1.234 µm	290 min	A3								
	454.6 nm	42.9 *	1748	290 %	\$3	157 rm	150.7*	711.3 nm	290 mm	\$3								1
	24.38 µm	155.9 *	0	100 %	0.4	24.38 µm	155.9.*	38.9 µm	290 min	14					51 mra	d		c
	0000000000				D4	20022				D4						_		
					64					84					/ Total Output	\/~Wobbing	<u>)</u>	107
					0.4													
					~					1000					Name	[mA]	Name	
					CS					C5					HP1	129.705	DP22x	Z
					CS A5					1000					HP1 HP2	129.706 133.722	DF22x DF22y	7
		Time s	tep 4h 🛨	Color for At	AS	<4h	<0h	<12h	< 16h	C5 A5	2 19h		_		HP1 HP2 DPH1x	129.706 133.722 2.495	DF22x DF22y QFx	7 2 12
0	orrection V %	Time : fanual correction		Colorfor &	AS	<4h	<8h	<12h	<16h	C5 A5	2.16h		_		HP1 HP2 DPH1x DPH1y	129.706 133.722 2.495 -4.332	DF22x DF22y QFx QFy	7 2 12 18
		fanual correction	۲.		A5	<4h	< 8 h	<12h	< 36h	C5 A5	¥16h	1			HP1 HP2 DPH1x DPH1y DPH2x	129.706 133.722 2.495 -4.332 -9.357	DF22x DF22y QFx QFy HFx	7 2 12 18 0
0	onection V is				AS	<4h	<0h			C5 A5					HP1 HP2 DPH1x DPH1y DPH2y DPH2y	129.706 133.722 2.495 -4.332 -9.357 4.120	DP22x DP22y QPx QPy HFx HFy	7 2 12 18 0 0
0		fanuel correction	۲.	arse F	A5	x4h	< 8 h	c12h C3		C5 A5				_	HP1 HP2 DPH1x DPH1y DPH2x DPH2y DPH2y DP11x	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187	DP22x DP22y QPx QPy HPx HFy IShx	7 2 12 18 0 0 11
0	100 %	A1	Atoo	arse 7	A5	x4h	<0h			C5 A5					HP1 HP2 DPH1x DPH1y DPH2x DPH2y DP11x DP11y	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -75.230	DF22x DF22y QFx QFy HFx HFy IStx ISty	7 2 12 18 0 0 11 21
0	100 % E2	A1	Ateo Alteo	arse 7	A5 ocus t-2nd	x4h	<8h			C5 A5					HP1 HP2 DPH1x DPH1y DPH2x DPH2y DPH2y DP11x	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -15.230 -4.726	DF22x DF22y QFx QFy HFx HFy IShx IShy DShx	7 2 12 18 0 0 0 11 21 -5
	Reset 82	Al A2	Ateo Alloo	arse 7	A5 ocus t-2nd	<4h	¢9k			C5 A5				ing jack	HP1 HP2 DPH1x DPH1y DPH2x OPH2y DP11x DP11y DP12x	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -75.230	DF22x DF22y QFx QFy HFx HFy IStx ISty	7 2 12 18 0 0 11 11 21 -5 -18
	E2 Reset B2 L connect i nt-Configu	Al A	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		cOh			C5 A5			<b>Г</b> 5	emil Lock	HP1 HP2 DPH1x DPH1y DPH2x OPH2y DP11x DP11y DP12x DP12y	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -15.230 -4.726 -3.412	DF22x DF22y QFx QFy HFx HFy IShx DShx DShy DStx	7 2 12 18 0 0 11 11 21 -5 -5 -18 10
	E2 Reset B2 L connect i nt-Configu	Al A2 Fine A3 cn establish retion has 4-18	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		<0k			C5 A5			-	imit Lock Geer Log	HP1 HP2 DPH1x DPH2x DPH2x DPH2x DP11x DP11x DP12x DP12x DP12x DP12x DP13x	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -15.230 -4.726 -3.412 2.747	DF22x DF22y QFx QFy HFx HFy IShx DShx DShy DStx	jmA 7 2 12 18 0. 0. 11. 21. 5 48 10. -2 554
	L connecti Reset B2 I connecti nt-Configu R Farmel v	Al A2 Fine A3 cn establish retion has 4-18	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		<0k			C5 A5					HP1 HP2 DPH1x DPH2x DPH2x DPH2x DP11x DP11x DP12x DP12x DP12x DP13x OP13y	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -15.230 -4.726 -3.412 2.747 -1.245	DF22x DF22y QFx QFy HFx HFy ISitx DSitx DSitx DSity DSitx DSity	7 2 12 18 0 0 0 11 21 21 21 55 4 8 10 22 554
	L connecti Reset B2 I connecti nt-Configu R Farmel v	Al A2 Fine A3 cn establish retion has 4-18	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		<0k			C5 A5				GearLog	HP1 HP2 DPH1x DPH2x DPH2y DP11x DP11y DP12x DP12y DP13x DP13y QP_FLx	129.706 133.722 2.495 -4.332 -9.357 4.120 -24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000	DF22x DF22y QFx QFy HFx HFy ISitx ISity DStx DSty DSty LarL	7 2 12 18 0 0 0 11 21 21 -5 -18 10 -2 554 79
	L connecti Reset B2 I connecti nt-Configu R Farmel v	Al A2 Fine A3 cn establish retion has 4-18	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		<01			C5 A5				GearLog	HP1 HP2 DPH1x DPH2x DPH2y DP11x DP11y DP12x DP12y DP12x DP12y DP13x DP13y QP_FLx QP_FLy	129.706 133.722 2.495 -4.332 -8.357 4.120 -24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000 0.000	DF22x DF22y QFx QFy HFx HFy ISitx ISity DStx DSty LorL TL12	7 2 12 18 0 0 0 11 11 21 21 -5 -5 -18 10 -2
	L connecti Reset B2 I connecti nt-Configu R Farmel v	Al A2 Fine A3 cn establish retion has 4-18	Alco Alco S	arse fr arse Is 3 A	A5 0cus 2+2nd 3+S3		<0k			C5 A5				GearLog	HP1 HP2 DPH1x DPH2x DPH2y DP12x DP11x DP12y DP12x DP12y DP12x DP12y DP13x DP13y dP_FLx dP_FLy dP_FLx	129.706 133.722 2.495 -4.332 -8.357 4.120 -24.187 -15.230 -4.726 -3.412 2.747 -1.245 0.000 0.000 0.000	DF22x DF22y QFx QFy HFx HFy ISitx ISity DStx DSty LorL 1L12 TL21	7 2 12 18 0 0 11 21 21 -5 -18 10 -2 554 75 554 120

- 7. Select Tableau type to "Standard", change Outer Tableau tilt [mrad] to 22 (or 24). Press **Start**. If the parameters except C1 is larger than Confidence, press **Accept**.
- 8. The program will automatically enter correction page. Select correction power (50% ~ 100%) and press the parameter (S3, B2, A2) that you would like to correct. Choose 100% correction power for lower order operation, like A2 and B2. Choose 50% correction power for higher order operation, like S3. Then press shortcut to start Tableau measurement again, repeat Step 7 to 8 until the parameters are less than or close to confidence values.

Note: Order of correction (low order operation to high order of operation) is C1, A1, B2, A2, S3. Note: if after Tableau measurement, the value stays the same but the angle changes, which indicates overshoot. If this happens, lower correction power in correction page and remeasure in Tableau.



#### **EDS** operation

TITAN Themis has a SuperX detector installed for energy dispersive x-ray spectrum collection. When doing EDS, use the double tilt holder and load the TEM grid with sample side facing down (e.g., the side with sample applied on).

- 1. Before switching to STEM EDS mode, make sure that the CCD camera is retracted and <u>fluorescence screen is lowered</u>.
- 2. Load a EDS setup in FEG Registers, e.g. "EDS 200 kV".



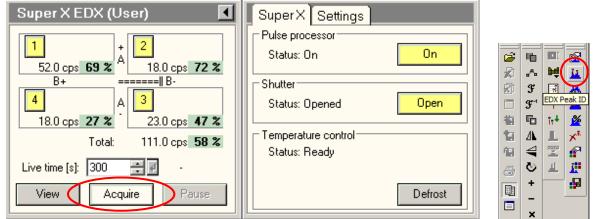
3. Click **Search** in **STEM**>**STEM** imaging to get an image in TIA. (Insert HAADF detector in **STEM** tab of UI if it's not inserted.) You may need to lower magnification to

STEM Ima	iging (Exper	rt) 🕨
STEM	Rotation (*)	: 0.0
Enable LM:	scan <u>90&lt; 1</u> <	0 >1 >90
	Dwell time (µs):	10.4
Blank	Scan frame: Pixel size:	512 x 512 291.4 pm
Search	Preview	Acquire
Focus	Scope	Auto C/B



find sample. Adjust Z height to bring the image close to focus. Fine adjust **Focus** to make the image more in focus.

4. (Optional) For quick EDS measurement, open Super X EDX (User) in UI, move the beam position marker to where you want to collect EDS spectrum and start Acquire. Press EDX Peak ID to get elemental analysis. Stop Acquire after getting enough counts.

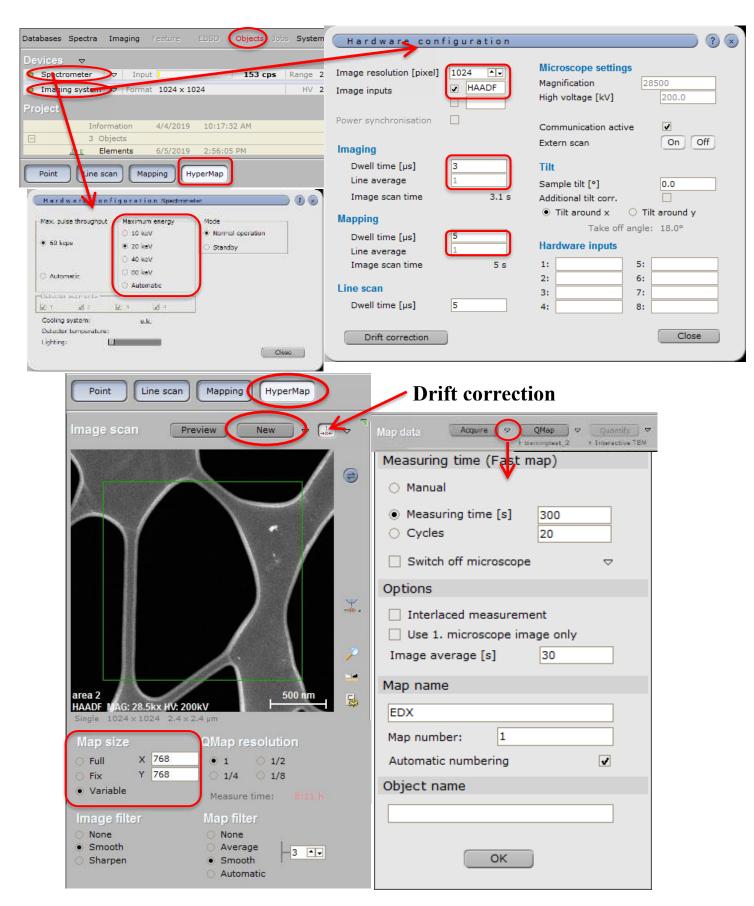


5. For advanced EDS measurement, open Esprit software. Start the Esprit software by double clicking desktop icon or by clicking icon in the Windows quick launch panel, log in with default username and password.

Start	6							斟	1
		User	edx						
		Password	***						
		Server	Loca	l server		•	Login		
			V S	ave pass	word				

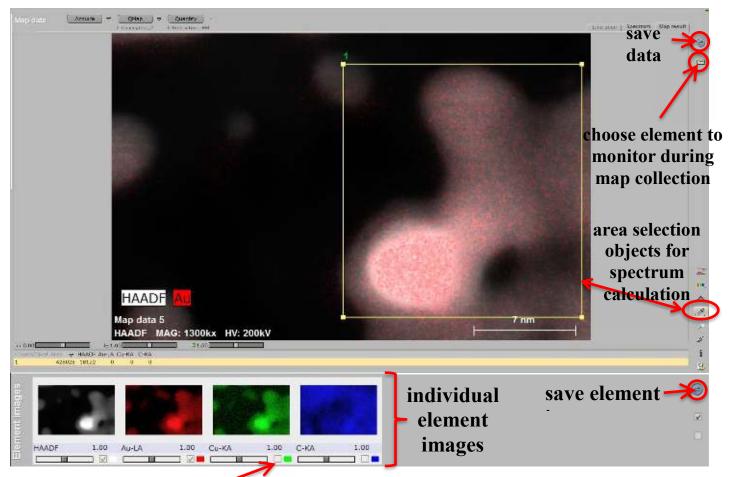
- 6. Select **Objects>HyperMap** in workspace, set up energy range (20 keV is good for most application) in **Spectrometer** page and resolution (1024), detector (HAADF), Mapping dwell time (5-20 us) in **Imaging system** page.
- 7. Click **New** in **Image scan** panel to obtain a STEM reference image. Drag the corner of the box to select area to collect fast map and activate Drift correction button. Note: there are rare occasions when the reference map cannot be collected but you can still collect map. Restart the computer will solve the problem. Ask EM staff for help. Note: there are three methods to define map size.
- 8. Click the triangle icon to the right of Acquire button to set up acquisition time. Manual: acquire EDS hypermap until user clicks Stop. Measuring time: set up total acquisition time. Cycles: set up number of frames to acquire and integrate for EDS hypermap. Note: during data acquisition process, clicking Stop once will terminate acquisition after the current frame and double-clicking Stop will terminate acquisition immediately. Note: reducing the gun lens number and beam spot size number can increase the beam intensity, thus enhances the x-ray counts per second.





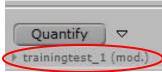


- 9. Click **Acquire** to collect hypermap of selected area. A status bar with remaining time is displayed in the upper right corner of the software interface. Select elements (multiple selection) from flap-out periodic table to monitor hypermap acquisition progress.
- 10. Save hypermap database and individual element images.



# display check box

- 11. Use area selection objects to select full or partial map for spectrum calculation and click Spectrum tab to show spectrum.
- 12. (Optional) Use **Quantify** to calculate the weight or atomic percentage of selected elements: click the method in grey under Quantify button, check Element identification, you can choose "Automatic" or select elements using "Preset list". Click **Quantify** button to calculate the elemental percentage.



Note: you can check Deconvolution ("Dec.") of some elements to exclude them in the final calculation.







#### CUNY ADVANCED SCIENCE RESEARCH CENTER

#### **Selected Area Diffraction operation**

1. In TEM mode, center the area of interest in the CCD image. Insert a Selected Area Aperture with preferred size (e.g. 40 or 10), center the aperture by activate Adjust and use MF X/Y.

Condenser 1	2000 💌	Adjust
Condenser 2	150 💌	Adjust
Condenser 3	Manual	
Objective	[none]	Adjust
Selected Area	20 €	Adjust

- 2. <u>Make sure the fluorescence screen is lowered</u>. Press **Diffraction** button on the right control pad to enter diffraction mode. Adjust Camera length and Focus to get sharp diffraction pattern in proper size.
- 3. Insert the beam-stop. Move the diffraction pattern by **Direct Alignments>Diffraction** alignment using MF X/Y to get the central diffraction spot blocked.
- 4. Lift the fluorescence screen and use CCD camera to take the diffraction pattern image (exposure time 0.5s or less).

Note: you can tilt the sample in Alpha and Beta angles to align it with desired zone axis, after that, readjust Eucentric Height.

5. When finishing, put down the fluorescence screen, retract the Selected Area Aperture and the beam-stop. Return to TEM mode by pressing **Diffraction** button.



#### Nanobeam Diffraction operation

- 1. In TEM mode, locate the area of interest in the CCD image.
- 2. <u>Make sure the fluorescence screen is lowered</u>. Load a NBD setup in FEG Registers.



3. Insert HAADF detector in **STEM** tab of UI. Click **Search** in **STEM>STEM imaging** to get an image in TIA. You may need to reduce magnification to find sample. Adjust **Z** height to bring the image in focus. Fine adjust **Focus** to make the image more in focus.

STEM Imaging	(Expert)	
OTENT.	Rotation (*):	0.0
Enable LMscan	<u>(0)&gt;1&gt;00</u>	>1 >90 0.4
100		12×512
Blank		191.4 pm
Search Pre	eview Ad	cquire
Focus	cope Au	ito C/B

- 4. Stop the Search in STEM, and put a beam position marker to where you want to take Nanobeam diffraction pattern.
- 5. Insert the beam-stop. Move the diffraction pattern by **Direct Alignments>Diffraction** alignment using **MF X/Y** to get the central diffraction spot blocked.
- 6. Retract the HAADF detector in STEM. Lift the fluorescence screen and use CCD camera to take the diffraction image (exposure time 0.5s or less).



ector (User)	
📑 320 mr	m
ctors	
/Brightness of:-	HAADF 💌
45.520 %	MF knobs
42.501 %	Auto C/B
	320 mi ctors /Brightness of:- 45.520 %

Note: you can tilt the sample in Alpha and Beta angles to align it with desired zone axis, after that, readjust Eucentric Height.

Note: you can use **Experiments** to acquire Spectrum image in a matrix. **Add markers** to the region where to take Nanobeam diffraction patterns and press **Acquire** to start. When finish, save the data base. Put an "Image position marker" on the STEM image, press "Ctrl" + "Alt" + "Shift" and drag the diffraction image to the image position marker to link them.

	Experiments		3	Settings		
	Select component:		E	Acquisition settings		
	SpectrumCollection 🔹	Ш		Image size (X)	10	
	Select experiment:	Ш		Image size (Y)	9	
				Fixed direction	X	
	Spectrum image			Dwell time (ms)	200	
		Ш	E	Configuration settings		
				Acquire EDX spectra	No	
				Acquire Cod images	Yes	
1	Add markers A Acquire			Acquire STEM image(s)	No	
		Ш		Elemental processing	No	
	Refresh Re-acquire			Post Beam Blank	No	
				Post Close Col. Valves	No	

7. When finishing, put down the fluorescence screen and retract the beam-stop. Return to TEM mode by loading a TEM setup in FEG Registers.



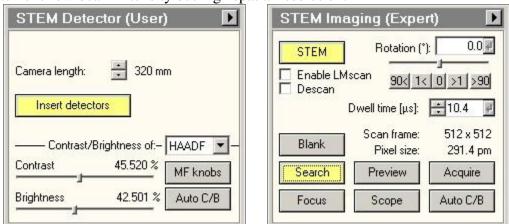
## **STEM Alignment**

Start from TEM mode (microprobe)

- 1. Locate area of interest in TEM mode. Finished the TEM alignments. (Cs image corrector is not necessary).
- 2. Bring the sample to Eucentric height using one of the three methods (step 2 in TEM Alignment).

Alignment in STEM mode (nanoprobe)

3. Make sure the fluorescence screen is lowered. Load a STEM FEG register. Note: default STEM mode is "Diffraction". Note: Larger spot size number and smaller C2 aperture aim to achieve low beam intensity but high spatial resolution.



- 4. Choose HAADF detector in STEM tab of UI. Set camera length to 210 mm. Click **Search** in **STEM>STEM imaging** to get an image in TIA. You may need to reduce magnification to find sample. Adjust **Z** height to bring the image in focus.
- 5. Center an area that is not region of interest (ROI) but contains some features (This region will be sacrificed in alignment). Check if there is condenser stigmatism: click Focus in STEM>STEM imaging to focus on a small area, Using Focus knob (you may need to use large focus step, e.g. 5) to wobble on both sides of focus to see whether there is image stretching. If yes, activating Condenser Stigmatism in Stigmator panel and using MF X/Y to correct condenser stigmatism.
- 6. Stop **Search**. Put a "Beam Position Marker" on the image. Double click the Beam Position Marker and set its position to 0,0 nm (uncheck "display in pixel first"). Park beam at amorphous carbon area to avoid damaging area of interest by beam.
- 7. Click "Diffraction" on the right control pad to enter Imaging mode.
- 8. If the beam (probe) is not visible, reduce magnification to find the beam, center it with **Beam Shift** in **Tune>Direct Alignments (MF X/Y)**.
- 9. Increase magnification to SA ~500 kx and center beam with Beam Shift (MF X/Y). If the probe is not circular. Make it circular by adjusting Objective Stigmatism (obj stig) in Stigmator panel. Sometime you need to use Focus knob to focus beam and then adjust obj stig. Note: this assumes that condenser stigmatism has been corrected in Step 5.
- 10. Center condenser (C2) aperture (50 um): Expand the beam with **Focus** knob to check whether the beam expands concentrically. If not, adjust condenser aperture (C2) so that



the beam expands concentrically when changing **Focus** knob. Note: In STEM mode, **Focus** knob is linked to intensity and **Intensity** knob is not functional.

- 11. Over focus (turn **Focus** knob clockwise to expand beam) the beam (probe) to see caustic image which contain a bright beam dot (hot spot) within halo. Note: You may not see the hot spot in the center, what you normally see is that certain part of halo edge is brighter than other area, this is ok since you will center the hot spot in step 13.
- 12. Click Rotation Center (Intensity) in Direct Alignment. Stop beam wobbling by turning Focus Step knob counter-clockwise.
- 13. Adjust **MF X/Y** to center the hot spot within the halo. Note: this step assumes that C2 aperture is centered in step 10.
- 14. Focus the beam to a spot (a spot with high intensity within the halo). Defocus can be reset by R2 button. Adjust **Beam tilt pp X/Y** in **Tune>Direct Alignments (MF X/Y)**: align the bright tips of the two images.
- 15. Center beam with **Beam Shift** in **Tune>Direct Alignments** (**MF X/Y**) if necessary.
- 16. Using **Focus** knob to shrink/expand beam on both sides. While doing so check the following:
  - a. Beam expands concentrically (C2 aperture is centered);
  - b. Hot spot is in the center of the halo (rotation center);
  - c. Objective stigmatism is corrected (probe to be circular).
  - If not, repeat step 9 -15.

Fine Tuning and Imaging

- 17. Click "Diffraction" on the right control pad to return to diffraction mode. Park the beam position marker away from ROI (usually at one of the four corners)
- 18. Click "Search" in STEM imaging, press "Eucentric Focus" and bring the sample to focus using Z height control
- 19. Watch the STEM image, carefully adjust focus and condenser stigmatism to achieve the sharpest image (no image stretch when changing focus on both sides), see step 5 for details.
- 20. Acquire STEM images.

Extra: Using Ronchigram to Adjust Focus and Condenser Stigmatism

- 1. After step 18 find an amorphous area and center it in the frame, stop **Search**, and park the Beam Position Marker at the center of the frame. See step 6 for details.
- 2. Change Camera Length to 510 mm. Watch Ronchigram on the screen. If Ronchigram is not centered and might be partial blocked by HAADF detector, using MF X/Y to center the Ronchigram.
- 3. Focus and carefully correct condenser astigmatism with condenser stigmator: use **Focus** knob to wobble on both sides of focus and minimize any stretching inside Ronchigram. Note: when showing Ronchigram, Focus knob is to zoom in/out of sample images, zoom in from both sides of focus to blow-up-point to check condenser stigmatism (stretched image change direction from one side to another; bright disk circular in center with dark ring indicates good condenser stigmatism).
- 4. Change camera length back to 210 mm.
- 5. Click **Search** in **STEM>STEM imaging** to find the region of interest, focus, and acquire STEM images.