**Basic SEM Operation FEI Quanta 200F**

Please wear gloves only when changing stages

1. Make file on left computer, First Name & initial of last name (if you do not already have one)
	1. Do not store images on computer long term
2. Home Stage (Shift F3)
	1. Menu Bar🡪 Stage🡪 Home Stage
3. Vent to open chamber
	1. Beam Control tab🡪Vent🡪Yes
4. Place stubs (samples) on stage
5. Select Vacuum level:
	1. High Vacuum – conductive samples
	2. Low Vacuum – non-conductive samples
	3. ESEM – must have staff assist for this mode
6. Close chamber door then click on “Pump”
7. Select Accelerating Voltage & Spot Size if needed
	1. Normal operation; 20kV, spot size 3
8. Wait for Vacuum status icon to turn green (both chambers) 
9. Manually move sample up to 10 mm line using wheel on mouse
10. “Beam On” – turns yellow when ready
11. Select upper left quadrant
12. Pause icon to start scanning – select lowest magnification possible (25x, 30x?)
13. F5 – toggle between 4 quad and 1 quad view
14. Adjust scan speed – use arrows next to turtle or rabbit
15. Photos/Imaging
	1. Choose magnification for imaging
	2. Increase mag
	3. Click on Reduced Area (focus box), focus image
	4. Click outside the box to return to normal view
	5. Return to lower/selected photo magnification
	6. Click on Camera icon to take Photo
	7. Save as: 8 bit Grayscale Image Files(\*.tif) – do not use 16 or 24 bit or program will not work correctly
16. End of Session
	1. Beam off
	2. Stage 🡪 **Home Stage**
	3. Vent 🡪 Yes
	4. Remove samples
	5. Shut door, select “High Vacuum”, click on “Pump” **wait** to be sure chamber has pumped out and is under vacuum before you leave
17. Return all settings to standard
	1. Spot size 3
	2. High Voltage; 20kV

 ETD mode

Standard Operating Procedure for FEI SEM

Do not attempt to use this instrument until you have been ***trained by FMIC staff***

**Protective Equipment**; Gloves when touching or handling anything inside sample chamber, or as needed when handling samples.

If you have any difficulties or problems please **ask for help** promptly

If you need a stage or detector other than one of the standards please make an appointment with FMIC staff prior to your schedualed FESEM time.

General use

* Turn on Computer screens (power button located on bottom of each screen)
* Right side computer is for the programs that operate the SEM\* (FESEM Control computer)
* Left side computer is for saving images (FESEM Image storage computer)

**Warnings:**

\*Do not save images to the FESEM Control computer this is only for operating the FE-SEM user interface programs.

**\*If you cannot send images to your folder, and you cannot find help, Restart the image computer, wait for it to boot up and reconnect, then try again. If it still won’t save, then save images to “Temporary” folder on desktop of FESEM Control computer and let a staff member know what you did.**

\*Do not attach any device to the FESEM Control computer, if this computer gets a virus the entire microscope will be unusable.

Note: **Always wear lint /powder-free clean room gloves when changing sample holders or detectors to minimise oils, dust, or other contaminants left inside the chamber**.

**Right screen**

 Look for "xTmcroscope Control /User Inter face" (UI)

 Click on: **Start**

**Log on**

Username: **supervisor** Password: **supervisor**

(both are lowercase)

The Image window should now be on the screen. It has 4 independent windows called 'quads',

to activate a quad click in it. Information bar at the bottom will be blue if the window is active.

Go to the **Stage** in the menu bar and select **Home Stage**

Right control Panel on right screen

Click on "**Vent**" to open SEM chamber. You will be asked to confirm this action.

 

Open the chamber door after the pump stops.

Place sample firmly in sample hole (push down to set firmly). Depending on the stage you may need to tighten the set screw.

Hold door shut and click on **Pump** to activate vacuum (button will be yellow when on). Select the appropriate vacuum mode (High vacuum, Low vacuum or ESEM). Hold the door shut until you hear the vacuum come on. It will be loud at first.



 

**Vacuum Status**;

**Raise sample to appropriate working distance.**

Move curser to lower right quadrent.

Hold down “ball” of mouse shile sliding mouse forward, continue holding until sample is at 10 mm line.

**DO NOT HIT COLUMN WITH SAMPLE**

**Activate the Beam**;

Click on Beam On (Right control Panel on right screen) –button turns yellow when active



 Click on a quad to activate it and then click on the pause button to start scan

Click on the sample chamber quad (lower right quad) to activate it, then click the pause

 button on the toolbar (or **F6)** to see the live image in that quad.

To move the sample holder up to the 10 mm mark;

- push down on the mouse roller ball and drag mouse up in sample chamber quad until sample is even with 10 mm mark on screen.

Push **F5** key to toggle between quad view and enlarged active window view

**Moving the sample**;

Double click on area of interest to center it, or

use arrows on keyboard or

Push down on roller ball on mouse and drag until sample is where you want it

**Changing Magnification**

There are several ways to change the magnification, choose what works best for you from this list;

* left click and draw box around sample
* "**+**" or "**-**" keys on keyboard to increase or decrease mag
* Select preset mag from box over upper left quadrant
* turn large knob on panel above keyboard
* use slider bar under magnification

**Focus**

Click on "Reduced Area" icon (or F7)

Use "Coarse" or "Fine" focus knobs on panel above keyboard or

right click mouse and move side to side

**Contrast and Brightness**

Adjust contrast and brightness knobs on panel above keyboard or

Videoscope icon or F3

**Stigmating**

**You need to correct astigmatism prior to taking images and when you change the imaging conditions (ie. increase/decrease kv, spot size, vacuum mode, detectors)**

1. Focus the image as well as possible.

 2. Bring the image just slightly out of focus. The image appears to become sharper in one direction whereas in perpendicular direction image blur increases (blurring or stretching of the image).

3. Bring the image just slightly out of focus in the other direction to observe the opposite directional blur.

4. Focus to the midpoint between the two directions, where the blur is visible.

5. Adjust image sharpness with the stigmator X and Y knobs, alternating each stigmator with fine focus knob until the best image is achieved.

**Taking Pictures**

1. Find area of interest, adjust contrast and brightness.

2. Increase magnification at least 1 - 2 times higher than final image magnification.

3. Click on "**Reduced Area**" Icon

4. Focus with "**Coarse**" then "**Fine**" knob (or just fine knob if already close to focus).

5. Stigmate with "**Y**" then **Fine focus**

6. Stigmate with "**X**" then **Fine focus**, (repeat stigmation & fine focus steps if needed)

7. Click on **Camera Icon** or **F2** button, wait for scan to be complete, save window will come up.

8. “Save As type” should be “Tif 8bit Grayscale Image Files(\*.tif)”

9. Confirm that "Save Image with Data Bar" is checked.

10. Confirm “Save Image with overlayed graphics” box is checked.

11. Make sure you save to the imaging computer (left computer), if you are not sure or it won’t automatically go to the imaging computer ask for help.

**Changing Samples** (Move to new sample or Replace samples in chamber)

Move from one sample to next on 7 position multi-stage;

1. Select Navigation tab

2. Double click on desired position number – sample should move down and then over to correct position.

3.Raise sample up to 10 mm mark

**Remove or Replace samples in chamber**

Click on “Pause “ icon to stop scanning

Click “Beam” to turn off “Beam off

Vent

Under “Stage” in menu bar select “Home Stage”

Once vacuum is released you may open door and remove your samples

**Shut down**;

Remove your sample(s).

**ALWAYS LEAVE CHAMBER UNDER HIGH VACUUM**

If FE SEM has been set up for Environmental Mode, low vacuum mode, backscatter or etching please return it to standard set up before you leave.

Standard Setup;

 Microscope;

* + - 1, 2 or 7 sample holder stage
		- backscatter detector removed
		- home stage (ask if you don’t know how to do this)
		- **door closed & chamber pumped out to High Vacuum**
		- aperture at 5 – do not change aperture on your own, always ask for help

 Microscope Control Computer (UI)

* all detector windows reset to ETD detector
* high vacuum mode
* spot size 3
* Set “High Voltage” to “20”
* turn off measurement tool and delete all measurements from window

 Other

* turn off picoammeter

If you have changed anything else (anything on the microscope itself, the imaging computer or the UI (user interface program) please change it back!

Put tools away

Return things (chairs, pencils, equipment) to original places

**Move chairs so they don’t block the door – it has an auto-close for fire protection**

**High resolution** - 50,000 x and higher

 For High resolution work It is important to degauss, align and stigmate the lens.

**DEGAUSS** (F8) 

 triggers the procedure which puts all currently used electron lenses to a normalized state by removing their hysteresis effects. For a few seconds while the procedure is running all live images disappear or turn fuzzy, and then return back. Use this function with (almost) focused image to obtain the most accurate Magnification, Horizontal Field Width (HFW) and Working Distance (WD) readouts.

**LENS ALIGNMENT** (Shift + F4)

The Lens Align. button starts automatic objective current oscillation (periodically under- and over focuses the image in a narrow range) to facilitate the process. Try to bring the rotation center to the screen center (if the magnification is too high, the rotation could seem like a linear motion instead of a rotation). When corrected, click the Lens Alignment toolbar icon to switch it off.

**CORRECTING ASTIGMATISM**

This optical aberration is caused by different focal lengths for rays of various orientation, resulting in a directional image blur (horizontal and vertical rays are not focused to the same plane on the image edges).

**You need to correct astigmatism when you change the imaging conditions.**

If astigmatism is severe and the cross is close to the edge of the screen when nearing correction, release the right mouse button, and reposition the cross in the center of the screen. Then repeat the procedure above to perform further astigmatism correction. You can use reduced area advantageously (see Chapter 4). If an astigmatism cannot be corrected, there may be some other reason, usually the final lens aperture is dirty (see Chapter 8), the magnification may be too high for the beam spot size (see below) or the sample is charging (apply conductive layer or use the LoVac / ESEM mode).

Step Action 1. Focus the image as well as possible.

 2. Bring the image just slightly out of focus. The image appears to become sharper in one direction whereas in perpendicular direction image blur increases (blurring or stretching of the image).

3. Bring the image just slightly out of focus in the other direction to observe the opposite directional blur.

4. Focus to the midpoint between the two directions, where the blur is visible.

 5. 1. Use the Beam module Stigmator 2D control. 2. The Mouse: press shift and hold the right mouse button pressed while in the active quad. This results in a 4 arrowed cross appearing on the screen with the cursor position at its center. Still holding the right mouse button down, move the cursor around the screen to achieve maximum sharpness. When you are satisfied, release the mouse button. 3. The MUI (optional): adjust image sharpness with the stigmator X and Y knobs until the best image is achieved. The computer beeps when the stigmator limits are reached. 6. Repeat steps 1–5 as necessary.

Additional Information;

**Specimen Preparation and Handling**  The specimen material for High Vacuum mode must be able to withstand a low pressure environment (without outgassing) and the bombardment by electrons. It must be clean and conductive. Oil and dust may contaminate the chamber environment, which could hinder or even prevent evacuation to the level needed for standard SEM operation.

 Note: **Always wear lint- / powder-free clean room gloves when reaching into the specimen chamber to minimise oils, dust, or other contaminants left inside the chamber**.

**NATURAL SPECIMEN**  If no coating is desired the Low Vacuum mode can be used to stabilise the specimen for observation. This mode is useful if there is a suspicion that a coating might alter the specimen. If the specimen contains any volatile components, such as water or oil, and therefore will not withstand coating, then the ESEM mode can be utilised with the correct environment gas and pressure to allow observation of the specimen in its natural state.

 **COATED SPECIMEN**  If the specimen is nonconductive (plastic, fibre, polymer or other substance with an electrical resistance greater than 1010 ohms) the specimen can be coated with a thin conductive layer. This conductive layer reduces beam instability due to sample charging and improves image quality. For successful imaging, rough surfaced specimens must be evenly coated from every direction. Biological, cloth and powder specimens may require carbon or other conductive painting on portions of the specimen that are hard to coat.

Coating reduces beam penetration and makes the image sharper. It may mask elements of interest for X-ray analysis (thus the use of carbon for geological and biological specimens).

LFD - Large field Detector - unrestricted field of view. Used for general imaging and gaseous SE detector, can be used simultaneously with BSED

GSED - Gaseous Secondary Electron Detector - general wet imaging and for high pressure imaging with gases. Pure SE singal, low mag range 240 at 7mm workiing distance. Best for resolution imaging. Used with Peltier stage and STEM stage

Best resolution in WetSTEM - 30Kv, spot size 1

 For more information on specific preparation techniques, see Scanning Electron Microscopy and X-Ray Microanalysis, 2nd ed. by Joseph Goldstein et al., Plenum Press, New York, 1992. MOUNTING THE SPECIMEN TO THE HOLDER Wafers and PGA devices have individual sample-mounting procedures. If you are using a wafer piece or other sample, attach the specimen to the specimen holder using any suitable SEM vacuum-quality adhesive, preferably carbon paint. **The specimen must be electrically grounded to the sample holder to minimize specimen charging**. If you are using a vice mechanism or double-sided tape, make sure the specimen is conductively attached to the holder. Note: The sample holder is not directly grounded to the chamber ground because it is connected to the BNC feed on the chamber door. This allows to measure the sample current.

 **Caution! Store samples and sample holders in a dry and dust free environment. Dust on samples can get drawn into the electron column, degrading imaging and requiring an FEI Customer Service.**



DO NOT CHANGE APERTURES – ASK FMIC STAFF FOR HELP

**No. Standard Recommended use (related to Standard sizes)**

**1** **1000** Service Alignment (hole in frame)

**2**  **- -**

**3**  **50 µm**  High current applications

**4** **40 µm**  X-ray mapping of low-Z elements at low voltages

**5** **30 µm**  **General imaging** or X-ray analysis

**6** **30 µm**  Dynamic experiments

**7**  **20 µm**  High resolution imaging

**Spot Size**





