PROCEDURES FOR OPERATION OF THE JEOL 5600LV SEM

PLEASE READ THROUGH THESE NOTES
(EVEN IF IT'S JUST TO REFRESH YOUR MEMORY!)

Amended October 2001

When the microscope is not in use, both PC monitors should be off, and both PCs should be on.

The microscope will be on (the Start key should be in the ON position).

As the microscope is kept under vacuum at all times, you will hear the pumps maintaining the vacuum. If there is silence - call the Technician.

As a general 'housekeeping' point - never touch the inside of the specimen chamber without gloves (cloth or plastic), also DO NOT TOUCH your specimen stub with bare hands.

ELECTRON BEAM GENERATION

1. Turn the microscope PC monitor on, and if you will be using the EDX equipment, turn this PC on too.
2. At the prompt, enter your network login, if appropriate. If not, click CANCEL.
3. At the desktop, click JSM5600 icon. The microscope will take 1 - 1½ minutes to initialise.
4. Ensure the Z control is 20 mm or greater- this can easily be changed later. If you have an extra large specimen to be viewed, increase the Z control to the maximum, 48mm.
5. Click SPECIMEN EXCHANGE followed by VENT to vent the chamber, or, you can use the orange VENT button under the microscope. Press this for a few seconds – it will start flashing. *Put the gloves provided on. DO NOT TOUCH THE INSIDE OF THE SPECIMEN CHAMBER WITH BARE HANDS.*
6. After about 30s (when the orange VENT light stops flashing), gently pull the front of the microscope. This may need a little extra pull, but don't force it! Insert specimen into holder, replace this on the microscope stage and close the front.
7. Whilst holding the front closed, either press the EVAC switch underneath the microscope for a few seconds, or click on EVAC in the SPECIMEN EXHCANGE menu. After a couple of seconds the microscope will start by rough pumping the vacuum - you will hear the pumps kicking in. If they do not, hold the stage door firmly closed and press the EVAC switch under the microscope again for a couple of seconds. If this does not work - call the Technician.
8. When the status box shows READY, close this window and click on HT. If you want to chose one of your pre-saved menus, click RECIPE (top right corner of screen) and choose the appropriate recipe.
9. If necessary, use auto contrast and brightness (ACB) and auto focus to gain an image.

The controls:
Most of the controls can be operated using either the PC or the console.
The specimen is moved using the X and Y controls on the front of the specimen chamber door.

Accelerating voltage (kV):	lower kV produces less signal but reduces charging and specimen damage. Chose between 10 - 15 kV initially. The lower the kV, the better.
Magnification:	limited by the spot size and screen resolution so that above x20,000 there is no more detail - just a bigger picture
Working distance (WD):	the distance between the final lens and the specimen surface. It is altered using the Z control and then focussing the image. A long WD gives good depth of focus, and vice versa. A short WD gives increased resolution. Use as short a WD as your specimen will allow.
Spot size:	small spot size gives increased resolution but less signal (therefore less charging, but a poor signal:noise ratio). A large spot size is needed for X-ray analysis.

N.B. Signal:noise ratio: at fast scan speeds the beam spends less time at each point on the specimen producing less resultant signal. At slow scan speeds more signal is detected. The noise inherent in the system is the same at all scan speeds, therefore at a slow scan there is a better signal:noise ratio and therefore a clearer image.

Signal:	secondary electron image (normal topographical image) backscattered electron image (atomic number contrast image - areas of elements with differing atomic numbers are seen)
Scan 1	reduced area fast scan, easier for focussing and stigmatism correction.
Scan 2	TV rate fast scan (image is refreshed 25 times per second), normal working speed producing a real time image
Scan 3	slow scan, improved signal:noise ratio producing a clearer image
Scan 4	very slow scan used for image capture

Contrast	)	On the PC screen, click on any of these, hold the mouse button and drag up or down to alter the parameter.
Brightness	)	
Focus	)	
Stig X	)	
Stig Y	)	

All of the above parameters, plus magnification, have a dial control on the console. It is generally accepted that you have more control using a dial than using a mouse, therefore I would advise using the console dial controls as much as possible.

If the image appears to shift diagonally when through focussing there is a stigmatism and it will not be possible to focus the image accurately. To alter the stigmatism select Scan 1 and alter Stig X and Stig Y to focus the image. Then select LENS RESET in the TOOLS menu and re-focus the image.

RECORDING AN IMAGE

There are two ways to record an image - by saving to a networked file, or by recording on photographic film. Using the digital storage method is a lot quicker, cheaper and easier. When using a photo quality printer there is not a huge difference in quality between that and photographic paper.

1. Pick the area of interest. Optimise the focus by selecting Scan 1, increasing the magnification and focussing on an edge. Decrease the magnification to the level wanted and select Scan 3. You can check the contrast and brightness levels using scan 3. This gives a better idea of how the image will look. If you need to adjust the focus, stigmatism or magnification, switch to Scan 2 to do so. To change the data on the information bar, open SETUP, select PHOTO DATA and you can change any of the options available. Press return after you have changed any text. Then press OK.
2. Select Scan 4. Wait until one full scan has completed. The information bar will show on the screen and the image will freeze.
3. Open FILE menu, select SAVE IMAGE FILE. *VERY IMPORTANT: mark box to merge text or the information bar will not be saved.*
4. Chose your folder OR if you do not have one, chose 'shared' folder on 'mann on mole'.
5. Name the file and press OK. It is saved as a bitmap file.

To record an image on film:

1. Follow steps 1 and 2 above.
2. Click once on the PHOTO button and keep clear of the microscope to prevent any vibration.
3. Once the photo has finished - hold the small black switch on the camera away from you while you pull the large film advance lever towards you once, and release. WRITE IN THE MICROGRAPH LOG BOOK THAT YOU HAVE TAKEN A PICTURE - THIS IS ESPECIALLY IMPORTANT AS THE MICROSCOPE DOES NOT AUTOMATICALLY COUNT THE NUMBER OF EXPOSURES.

USING THE X-RAY ANALYSIS EQUIPMENT

As in the TEM it is possible to use the X ray analysis to get an elemental spectrum. In the SEM, it is also possible to convert this into an elemental map showing areas of different elements. Make sure the sample for analysis is coated appropriately, e.g. when looking for calcium phosphate, for instance, do not coat in platinum because the platinum M line sits very near to the phospour K lines. In that instance you would use a carbon coating. However, if you are wanting to map areas of carbon in your sample, obviously do not coat in carbon, use platinum.

The X ray analysis probe is kept in position at all times in the SEM.

1. Chose the area that you want to map.
2. The kV and spot size will need to be increased to boost the signal and get sufficient readings to produce a spectrum. It is therefore recommended that any secondary electron image needed is recorded before analysis as this may cause damage to the specimen.

3. Select the Link ISIS icon on the microanalysis computer.
4. At the next screen, click on the Labbook. Then click on the icon that looks like a spectrum.
5. Change the kV to 20kV. Reduce the contrast and re-focus.
6. Ensure the working distance is at 15mm. Re-focus.
7. Click on Go.
8. Check the percentage dead time - this should be about 30%. If it is less than this, increase the spot size.
9. During acquisition, you can check the elements present by clicking on the question mark, then click on Auto ID. This will bring up a list of elements it thinks are present. Click on any of these and a line will appear on the spectrum telling you which peak corresponds to that element. Click on Label Peaks to identify all the peaks. If you want to know what an unidentified peak is (use the 'magnifying glass' button to increase the size and scaling of your spectrum) then right click on that peak. The software will tell you what it thinks the peak is, and a list of possibilities as well. If you highlight any of these, a series of blue lines will appear on the spectrum. These are all the keV emission values for that element. If these lines match your series of peaks then that element is present.
10. Spectra can be saved to the hard drive by selecting File, Save a Spectrum. Or they can be saved to the network by selecting File, Export as Tiff, m drive etc.

Using the Mapping function.

To produce an elemental map of an area:

1. Select the elements of a spectrum that you wish to see by right clicking on that peak followed by PAINT WINDOW. You can choose as many elements as you like to map.
2. Then click on the button that looks like a grid.
3. Click on GO and wait for the images to fully appear.
4. These map groups can be saved onto the hard drive. To transfer them across to the network, each elemental map has to be moved and saved individually. So, at the map group, highlight the elemental map you want saved (red diamond in the right corner), go to File, Export as TIFF, choose m drive and save as normal.

Spot Analysis using the Autobeam function.

1. Carry out steps 1 – 6 above.
2. Go back to the Labbook, click on the icon next to the spectrum (looks like a microchip!!).
3. Click on Go (triangle shape). This will lock you out of the microscope controls while the ISIS system is collecting an image.
4. Click on Stop (square shape).
5. Click on the spot on the image that you want t o collect from.
6. Go to the spectrum and click on Go. Correct the spot size to get adequate acquisition.
7. You can save the Autobeam image to the hard drive or to the network.

BACKSCATTERED ELECTRON IMAGES

Backscattered electrons are used to produce 'atomic number contrast' images. These are not topographical and the detector is positioned above the sample rather than to one side. For this reason, the best BSE images are produced from flat, polished surfaces. If you have a sample with an overall low atomic number, e.g. mostly carbon and you suspect the presence of heavy metal, a BSE image will show any areas containing elements with a high atomic number. Areas of high atomic number will appear brighter than areas of low atomic number. This is not a quantitative method of analysis, and it will not tell you the exact element present.

1. Turn the HT off.
2. Check the tilt is 0°. If using a very flat specimen, the minimum working distance is 8mm. If your specimen has any height, take this into consideration and increase the WD.
3. Insert the detector. Move the handwheel from position 3 to position 1. Hold the arm firmly as you will be manoeuvring the arm in and out of a vacuum. When inserting the detector the vacuum will try to 'suck' the arm in quickly - resist this and let it move in gently.
4. Open the SIGNAL box and select BEI.
5. At the BEI menu select COMPO. Select ACB as necessary to gain an image.
6. The image will be brighter at higher kV (15 - 25kV) and larger spot size, however more damage will occur. Also, use as short a WD as your sample will allow.
7. When you are finished, turn the HT off and pull the arm out to position 3.

LOW VACUUM MODE

The low vacuum mode can be used for semi-wet samples (for wet samples you need an ESEM), or samples that have not been coated. You are likely to see the effects of charging, however these will not be as bad as they would in a high vacuum. The pressure in the chamber is bought up to a maximum of 130 Pa - which is still less than atmospheric pressure.

It is possible to switch to low vacuum mode at any time by changing the MODE switch in the right hand corner. However, if you have a specimen that will be harmed if looked at in high vacuum mode first, the following procedure needs to be followed.

Using the LV if you have a semi-wet specimen:

1. Turn the microscope on in high vacuum mode and set the following parameters:

Acc voltage:	10 - 20kV
Spot size:	30 – ish
Working distance	greater than 10mm
Mag:	x5,000 max
Signal:	SEI

2. Switch to LOW VACUUM mode (right hand corner). The signal will change from SEI to BEI and the low vacuum adjustment panel appears.

3. Once the vacuum status becomes READY, set the pressure to between 50 - 70Pa and wait for that pressure to be reached.
4. Vent the specimen chamber to atmosphere, install the semi-wet sample and evacuate the chamber.
5. Once the vacuum status shows READY, insert the backscattered detector to position 1 (see previous page).
6. Click on the HT.
7. Adjust focus, contrast and brightness to obtain an image. You may need to use the ACB function.
8. Adjust spot size and chamber pressure to reduce any effects of charging. Increasing the chamber pressure will decrease the effects of charging, however you may need to increase the spot size to counteract any loss of signal. The maximum pressure which can be used is 130Pa.

SHUTDOWN PROCEDURE

When you have finished your session, it is important that you leave the microscope in a suitable state, ready for the next user.

1. Wind down the magnification to its lowest level.
2. Lower the working distance (Z control) to between 20 - 25 mm.
3. Ensure the spot size is between 20 - 25.
4. Turn the HT off.
5. Remove the specimen from the microscope using either the VENT and EVAC switches underneath the column, or the SPECIMEN EXCHANGE menu on screen, remembering to use gloves when handling the specimen/specimen holder.
6. *Always leave the microscope under vacuum.*
7. Close the JSM5600 programme, and log out. Turn the monitor off. Do the same with the microanalysis computer.