Olympus BX60M Microscope Standard Operating Procedure

Things to know BEFORE using the microscope

- Study the Quick Reference Guide to become familiar with the microscope.
- The U-LBD-2 filter is simply a green color filter that can you be used at your discretion for any of the observation modes.
- To "engage" a filter/slide, make sure it clicks twice when inserted.
- To "disengage" a filter/slide, simply pull it out from the "engaged" position, until it clicks once.
- To "remove" a filter/slide, take it out of the microscope. *
  *When a filter/slide is removed, put it in the designated Ziploc bag so it stays clean.
- When inserting filters/slides horizontally, make sure the label is facing upward.
- When inserting filters/slides vertically, make sure the label is facing forward.
- For additional information about the use of the microscope, refer to the Microscope Appendix.

Which observation mode should you use?

-Brightfield Illumination is the simplest technique and should be used when the sample already has good contrast.

-Darkfield Illumination is recommended for samples with small changes in topography.

-Differential Interference Contrast (DIC) is recommended for relatively thicker, opaque, or low contrast samples.

-Simple Polarized is recommended for anything with a crystalline structure.

STARTING UP

1. Take the cover off of the microscope.
2. Turn on the main switch on the back of the microscope.
3. Push the power button found on the monitor adjacent to the microscope.
4. Using the light intensity lever set the light intensity to at least 6.
5. To set up for the various observation modes, refer to the appropriate sections.

OBSERVATION MODES

Brightfield

1. Slide the cube selector knob to the BF position.
2. Uninstall the Nomarski Prism.
   a. Loosen the DIC clamping screw at the front of the revolving nosepiece.
   b. Disengage the U-DICR slide.
   c. Tighten the clamping screw to secure the prism.
3. Disengage the U-AN360 Analyzer.
4. Remove the U-PO Polarizer.
5. Engage the U-DND-2 Glare Shielding ND Filter in its place.
6. Engage the U-LBD-2 filter if desired.
7. Go to OBSERVATION METHOD.

Darkfield

1. Slide the cube selector knob to the DF position.
2. Uninstall the Nomarski Prism.
   a. Loosen the DIC clamping screw at the front of the revolving nosepiece.
   b. Disengage the U-DICR slide.
   c. Tighten the clamping screw to secure the prism.
3. Disengage the U-AN360 Analyzer.
4. Remove the U-PO Polarizer.
5. Engage the U-DND-2 Glare Shielding ND Filter in its place.
6. Engage the U-LBD-2 filter if desired.
7. Go to OBSERVATION METHOD.

Nomarski Differential Interference Contrast-Brightfield

1. Slide the cube selector knob to the BF position.
2. Engage U-AN360 Analyzer Slide.
3. Remove the U-DND-2 Glare Shielding ND Filter.
4. Engage the U-PO Polarizer in its place.
5. Turn the dial on the analyzer slide until you can no longer see anything on the monitor or the microscope. It should all look black. This is easily done at low magnifications (5x).
6. Install the Nomarski Prism.
   a. Loosen the DIC clamping screw at the front of the revolving nosepiece.
   b. Engage the U-DICR slide.
   c. Tighten the clamping screw to secure the prism.
      **Note- If an UMPlan objective is used push in the selector level. If an LMPlan objective is used, pull out the selector level.
7. Engage the U-LBD-2 filter if desired.
8. Go to OBSERVATION METHOD.

Nomarski Differential Interference Contrast-Darkfield

1. Slide the cube selector knob to the DF position.
2. Disengage the U-AN360 Analyzer Slide.
3. Disengage the U-PO Polarizer Slide/U-DND-2 Glare Shielding ND Filter.
4. Uninstall the Nomarski Prism.
   a. Loosen the DIC clamping screw at the front of the revolving nosepiece.
   b. Disengage the U-DICR slide.
   c. Tighten the clamping screw to secure the prism
5. Engage the U-LBD-2 filter if desired.
6. Go to OBSERVATION METHOD

Simple Polarized Light

1. Slide the cube selector knob to the BF position.
2. Engage U-AN360 Analyzer Slide.
3. Remove the U-DND-2 Glare Shielding ND Filter.
4. Engage the U-PO Polarizer in its place.
5. Engage the U-LBD-2 filter if desired.
6. Go to OBSERVATION METHOD.

OBSERVATION METHOD

1. Turn the revolving nosepiece to engage the 10X objective (be sure it clicks into position).
2. Using the coarse adjustment focus knob, move the sample stage all the way down.
3. Place your sample on the stage.
4. Put the light path selector knob in the middle position (be sure it clicks into position.)
5. Using the coarse adjustment focus knob, slowly move the sample stage up to bring the sample into approximate focus. (Be careful not to crash the objectives into the sample; they are very expensive to replace!!!)
6. Use the x-axis and y-axis knobs to move your sample so that it is in the field of view.
7. Use the fine adjustment focus knob to make any final adjustments.
8. Engage the objective to be used by rotating the revolving nosepiece.
9. If necessary readjust the focus and the light intensity.
10. Adjust the Field Iris Diaphragm (FS) and the Aperture Iris Diaphragm (AS).
    **Note-When using Darkfield mode, completely push in the Field Iris Diaphragm knob and the Aperture Iris Diaphragm knob.
11. To view different parts of the sample, move the stage by turning the x-axis and y-axis knobs.

SHUTTING DOWN

1. Move the stage all the way down.
2. Remove your sample
3. Reduce the light intensity lever to P.
4. Push the power button on the monitor adjacent to the microscope.
5. Turn off the main switch on the back of the microscope.
6. Put the cover back on the microscope.

Photomicrography

To take pictures of your sample refer to the Microscope Image Capturing Standard Operating Procedure.

**Note- The light intensity lever should be at least at the level indicated by the camera symbol, and the light path selection knob should be pulled out completely.

Microscope Appendix

Rotating the Stage

1. Using the L-shaped Allen screwdriver, slightly loosen the stage clamping screw found behind the sample stage.
2. Using the stage clamping screw, rotate the stage.
3. Tighten the stage clamping screw.

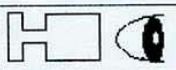
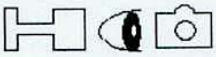
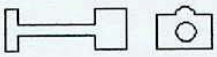
Stage Height Adjustment

Lower the stage to its lower limit. While holding the stage, use the Allen screwdriver found at the top of the microscope to loosen the clamping screw on the right side of the microscope. Adjust the stage to the desired height, and tighten the clamping screw.

Light Path Selection

The selector knob is ordinarily in the middle position.

**Note-When the Light Path Selector knob is completely pushed in you will not be able to see anything on the monitor.

Light Path Selector Knob	Symbol	Intensity Ratio	Application
Pushed in		100% for binocular eyepieces	Observation of dark samples
Middle position		20% for binocular eyepieces, 80% for TV/photomicrography	Observation of bright samples, photography, TV observation
Pulled out		100% for TV/photo-micrography	Photography, TV observation

Pre-focusing Lever

To put an upper limit on the coarse adjustment knob, turn the pre-focusing lever up. This makes refocusing easier when changing samples.

Using the Field Iris Diaphragm

Brightfield observation

For good image contrast, adjust the diameter of the illuminating beam in accordance with the objective in use. Adjust the diaphragm so that the field of view is circumscribed by the field iris diaphragm in order to exclude stray light.

Darkfield observation

Always keep the field iris diaphragm knob pushed in to leave the diaphragm open.

Using the Aperture Iris Diaphragm

Brightfield observation

In general, a good image is obtained if the diaphragm is stopped down to 70-80% of the objective's numerical aperture.

Darkfield observation

Always keep the aperture iris diaphragm knob pushed in to leave the diaphragm open.

Microscope Filters/Slides

U-LBD-2 Filter

Nomarski Prism

U-AN360 Analyzer

U-PO Polarizer

U-DND Glare Shielding Filter