BASIC OPERATION OF THE NIKON LABOPHOT 2A MICROSCOPE

This document describes the basic operating procedure for the Labophot microscopes when using them to examine opaque specimens. It starts by describing the procedure for bright-field reflected light microscopy and then it gives the basic procedures for polarized, dark-field and DIC imaging. Refer to figures 1, 2 and 3 if you have any difficulty finding any of the controls on this microscope. Tables 1 and 2 at the end of this document summarize that major specifications of these microscopes and figures 4 through 7 show examples of different imaging modes.

Note: These microscopes are configured for reflected light microscopy. The objective lenses are all bright field/dark field, plano, achromat lenses and they do not include corrections for cover slips. Four of the objective lenses on one microscope also include DIC capabilities.

Important: The objective lenses on these microscopes are all dry lenses, even the 100X objective. Using any immersion fluid can permanently damage them.

Precautions

1. Handle the microscope gently. Strong mechanical shocks can damage it.
2. When carrying the microscope support it by the bottom of the base.
3. Do not use the full power settings for the illuminators. This will significantly reduce the life of the bulbs.
4. Be very careful to never let the specimen touch the objective lens. This microscope has features which will help you avoid this. Use them.
5. Never use oil or any other immersion medium with this microscope. All objective lenses are "dry" lenses.
6. Never force any part. Doing so might permanently damage the microscope. If something is stuck, then it should be serviced. In other cases there might be another way to make the needed adjustment. For example, do not twist the camera to line up a feature on the specimen with the edge of the field of view. Rotate the stage instead.

Prepare the Microscope

7. Remove the cover and put it somewhere safe.
8. Inspect the microscope.
9. Turn on the epi-illuminator. Set the power to about 50%.

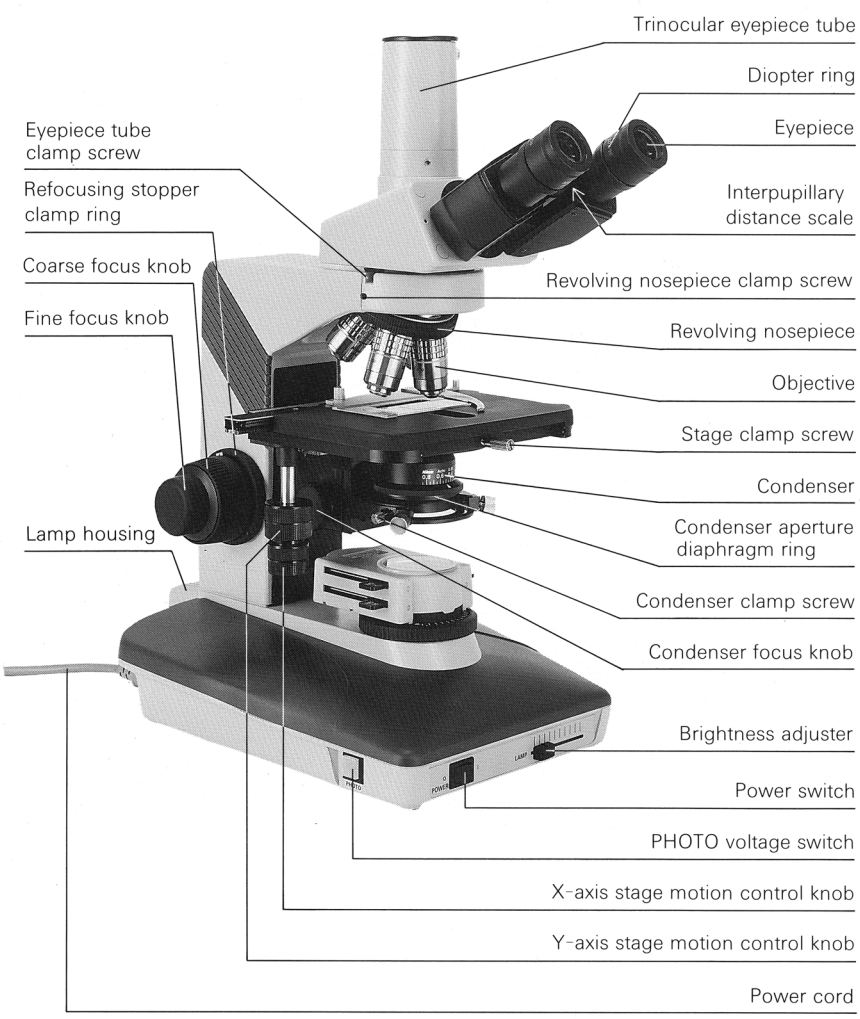

Figure 1. A view of the right side of the Labophot microscope. (From Nikon's instructions for the Labophot-2A.)

Prepare the Specimen

Care must be taken to prepare a specimen whose reflecting surface is perpendicular to the optic axis and to install it in the microscope without damaging the specimen or the microscope.

10. Mount the specimen on microscope slide. Use some modeling clay, lens paper and the Leitz press. Press only hard enough to get the specimen to stick to the clay and the square the specimen with the optic axis.
11. Release the refocusing stopper clamp.
12. Lower the specimen stage.
13. Select the 5X objective. This is the shortest objective and it has the longest working distance and the largest depth of field.
14. Install the specimen.
15. Raise specimen slide to within 2-4 mm of the objective lens, less than the working distance and yet a safe distance from the lens.

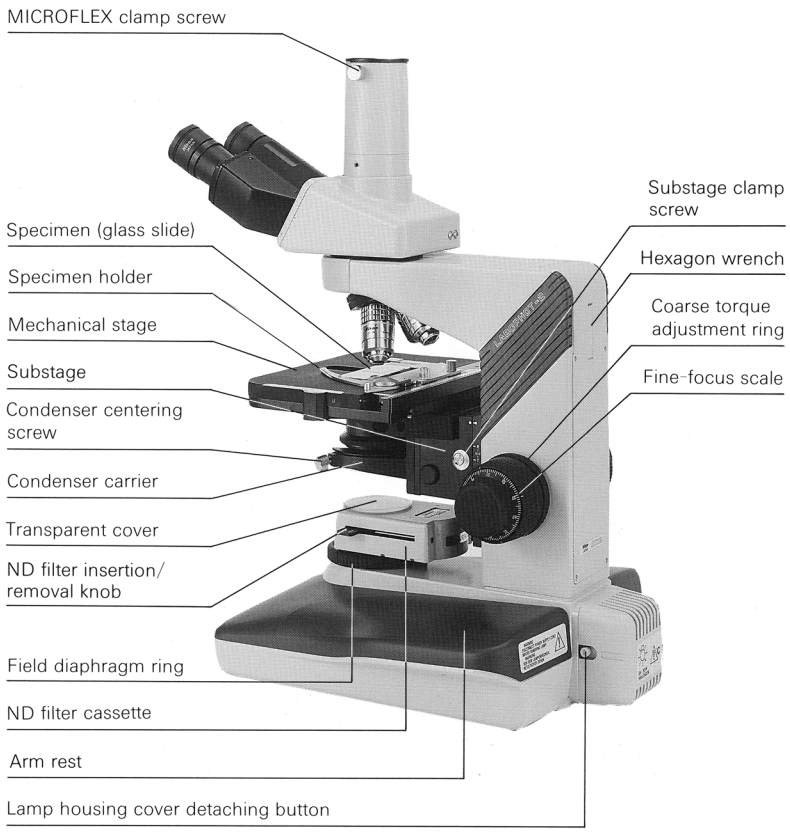

Figure 2. A view of the left side of the Labophot microscope. (From Nikon's instructions for the Labophot-2A.)

Initial Focus
This simple step includes a very important safety procedure.

16. Slowly lower the specimen until it comes into focus.
17. Set the refocusing stopper clamp so that you won't be able to crash the specimen into the objective lens.
18. Loosen the stage clamp screw and rotate the stage to orient the specimen to your liking. Tighten the screw again when done.

Note: the polarizer, analyzer and the Wolfstram prism, if present, may need to be removed or adjusted so that you can obtain a clear, bright image of your specimen.

Adjust Binocular Eye Pieces
Properly adjusted eye pieces are essential for working comfortably with the microscope and for obtaining the best image in both eyes.
19. Set the distance between the eye pieces so that you can comfortably view the specimen by

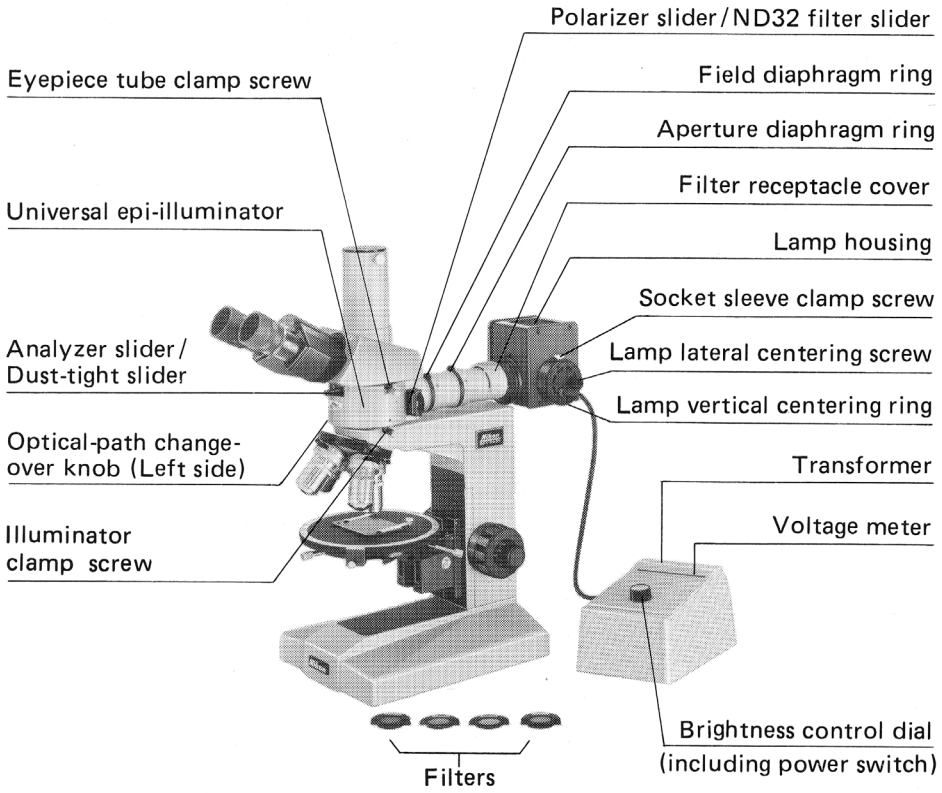

Figure 3. Identification of each part of the epi-illuminator. (From Nikon's instructions for the Labophot-2A.)

looking straight ahead using both eyes at the same time.

20. Close your left eye and focus the specimen using the fine focus knob.
21. Close your right eye and focus the specimen by turning the eye piece focusing ring.

Adjust the Illumination

A properly adjusted illumination system is necessary if you are to obtain the best resolution and contrast.

22. Open the field and aperture diaphragms.
23. Remove an eye piece.
24. Look into the eye piece's opening and observe the illumination of the specimen.
25. Stop down the field diaphragm until it blocks about 20% of the view of the specimen.
26. Replace the eyepiece.
27. Stop down the aperture diaphragm until it is just out of view.

Changing Magnification

The objective lenses are para-focal, meaning that they all focus on approximately the same specimen plane. After changing magnification only a slight adjustment of the fine focus knob will be necessary.

28. Rotate the objective lens turret to position the next higher/lower magnification objective in the light path.

29. Adjust the fine focus knob to obtain the sharpest image.

Note: It is a good practice to refocus each time an objective lens comes into the light path rather than skipping over one or more to get to the final magnification. Only slight focus adjustments will be needed for each step. But when skipping over objectives focusing will be more difficult because you will be so far out of focus and, if you are increasing magnification, depth of field decreases and this will require more precise focusing. Also, since working distance decreases with increasing magnification you will want to watch the objective closely to make sure it does not strike the specimen.

Polarized Light
Polarized light can aid in increasing the contrast in the image. This microscope employs two polarizing elements, a rotatable, removable polarizer and a fixed analyzer which can be removed from the light path but it cannot be rotated.

30. The analyzer can be in or out.
31. Rotate the polarizer to obtain the desired effect.

Dark Field Illumination
Dark field illumination can be used to image the details of the specimen and to ignore the larger features. It works by illuminating the specimen with a hollow cone of light. In reflected light microscopy the image is formed from light that is reflected from the edges of each feature, excluding light that is normally reflected from the flat areas.

32. Set the illuminator's power to maximum (on the last green LED, not the red LED).
33. Open the aperture and field diaphragms.
34. Remove the analyzer.
35. Pull the BF/DF knob out to obtain dark field illumination.
36. Focus.
37. Adjust the polarizer to brighten the image.

Note: Smooth specimens might not be good subjects for dark field imaging.

Differential Interference Contrast (available on only one microscope)
Differential interference contrast (DIC) can greatly enhance the quality of the image, bringing out the finer details in the specimen. It can also add striking but false colors and an illusion of depth.

38. Focus in the usual manner.
39. Make sure the analyzer is in.
40. Switch the Wolfstram prism to the "In" position.
41. Adjust the polarizer to obtain the desired effect.

Note: Only the objective lenses marked "DIC" are capable of differential interference contrast imaging.

Pulnix Video Camera
Preparing the Pulnix video camera involves simply turning it on and performing a white balance. Some of our cameras have a white balance button you can press. Most do not.

42. Start up Adobe Photoshop and select *file/import/plug-in digitizer*.
43. Remove the specimen from the microscope.
44. Turn on the illuminator for transmitted light microscopy. Set it to 50% power.
44. Pull the knob to divert the light away from the eye pieces and to the camera.
45. Turn on the video camera. If already on turn it off for a few seconds and then turn it on again. If it has a white balance button simply press the button.
46. Turn off the transmitted light.
47. Replace the specimen and refocus.
48. Stop down the aperture diaphragm until it is just out of view.

Polaroid Digital Camera

Preparing the Polaroid digital camera involves simply turning it on and using the correct TWAIN software.

49. Start up Adobe Photoshop and select *file/import/Polaroid DMC*.
50. Turn on the camera.
51. If using a PC to grab the images and the camera was off when the computer was booted you may have to rescan the SCSI bus. To do this open the control panel, start the *Tape Devices* program, click the *Detect* button and when the scan is finished close the *Tape Devices* panel.
52. Pull the knob to divert the light away from the eye pieces and to the camera.

When Finished

53. Position the 5X objective in the light path.
54. Lower the stage and remove your specimen.
55. Turn off the camera.
56. Turn off the illuminator.
57. Replace the cover on the microscope.

Figure 4. Wrought iron from a 19th century train. (Labophot 2A, 10X objective, bright field illumination.)

Figure 5. Wrought iron from a 19th century train. (Labophot 2A, 10X objective, polarized light.)

Figure 6. Wrought iron from a 19th century train. (Labophot 2A, 10X objective, dark field illumination.)

Figure 7. Wrought iron from a 19th century train. (Labophot 2A, 10X objective, differential interference contrast.)