



MICROSCOPE 1

Nikon SMZ1270 Stereomicroscope

TRAINING GUIDE

Instruction for use of Microscope 1. Detailing the techniques available, location of supplies, and requirements for use of this shared resource

[3364G KHS Advanced Microscope Suite](#)

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Introduction

Training requirements:

- Training on the use of this instrument is required prior to keycard access.
- First come, first served – if you are planning any time-sensitive analysis remember to reserve your time on the shared calendar to prevent any scheduling delays.
- Please remember to log your time, even retroactively, to provide data of usage.

Calendar information:

- Viewing of the shared calendar is accessible on the Microscopy Suite GVSU page
<https://www.gvsu.edu/clas/labresource/microscopy-facility-13>
- Access to the calendar is automatically added with Keycard request
- To add the calendar to your account please follow the steps outlined in “Advanced Microscope Suite Calendar Access”

Supplies available:

- Drawer 11 includes a ready supply of cleaning agents for the microscopes. If low, please email Ashley Vanhouten.
- Sparkle, IPA, Ethanol are the only cleaning agents approved for use in the suite.
- If there is an advanced issue please contact your PI, Aaron Perry, or Ashley Vanhouten for additional support.

If you encounter a situation where the microscope has become damaged or is malfunctioning in any way, please communicate this issue with your PI.

PIs, please communicate issues to Aaron Perry, Ashley Vanhouten (Equipment Repair), or Zach Hancock (IT Support) so we may provide support for this space. Examples of when to reach out include, but are not limited to:

- Bulb outages
- Software calibration issues
- Mechanical focus issues
- Error messages
- Initialization issues

Microscope Techniques

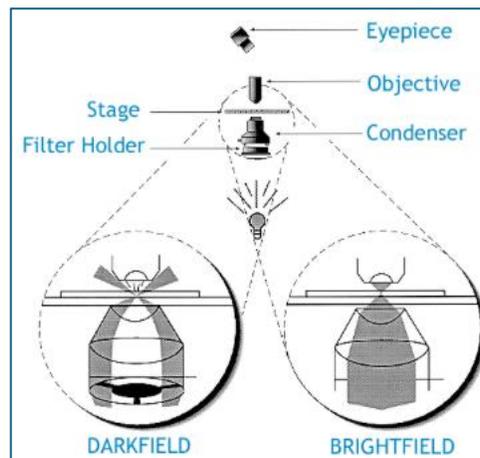
Bright-Field is the most basic microscopy technique and uses illumination that generates a path of light that will pass through your specimen. How the light path is modified (reflected-episcopic or transmitted -diascopic) as it passes through your specimen generates the image.

Reflected (Episcopic) Illumination versus Transmitted (Diascopic) illumination

Opaque specimens most commonly are best viewed using reflected light, while translucent and transparent specimens typically provide the best results under some variation of transmitted illumination. This is not always the case, other variables should be taken into consideration while forming a lighting strategy, including the following:

- Basic physical characteristics of the specimen (geometrical profile, topography, morphology, etc.)
- Type of information looking to extract from the examination of the specimen
- Photographing requirements
- How the information will be used

Dark-Field restricts the illumination (light path) in such a way that only the light that is scattered by your specimen is used to generate an image.



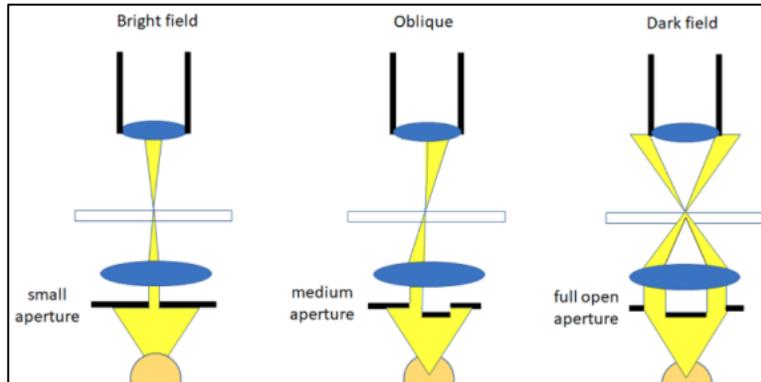
Using Darkfield Microscopy To Enhance Contrast:

An Easy and Inexpensive Method

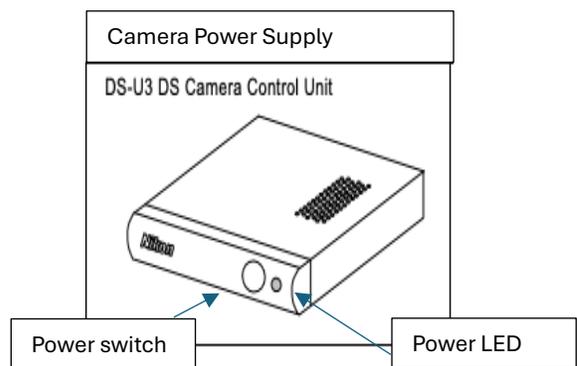
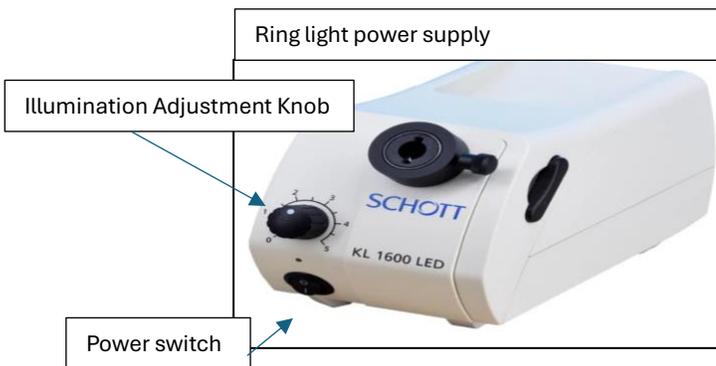
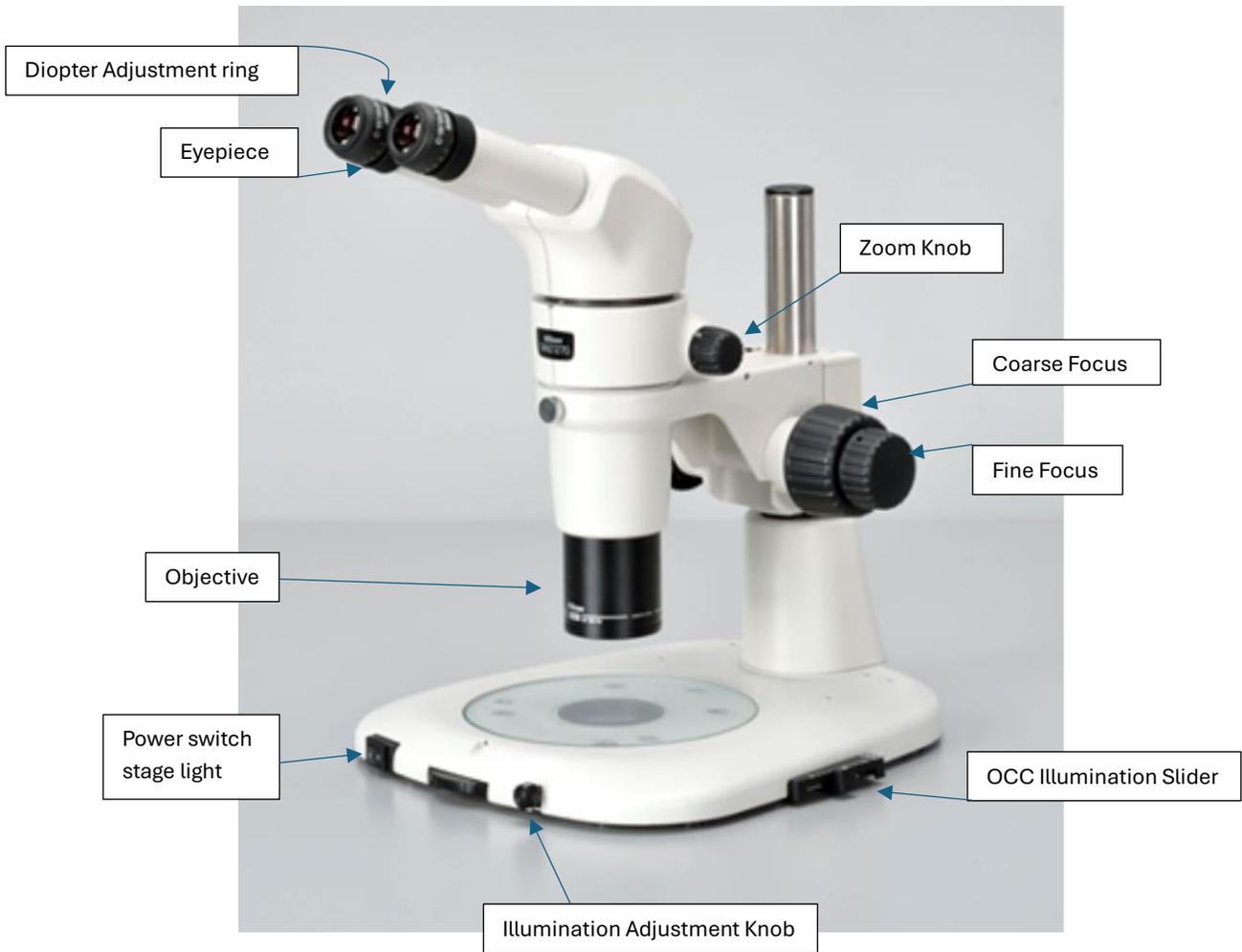
<https://public.wsu.edu/~omoto/papers/darkfield.html>

Oblique Coherent Contrast (OCC) illumination

The OCC illuminator utilizes a sliding diaphragm to optimize contrast in bright-field to dark-field illumination configurations. Utilizing the axial light (ring light) the diaphragm position can be moved to operate within Bright-field through various oblique settings to Dark-field microscopy. This can allow viewing of changes in the specimen appearance to highlight varying features.



Detailed Microscope Images



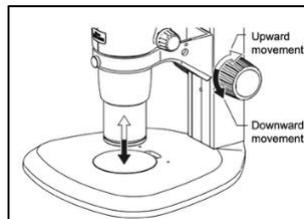
Bright-Field Adjustment

1. Power on light source – Diascopic illuminator (ring light/reflected light)
 - a. Power supply located to the left of the microscope
2. Verify optical path is set to eyes

Selector Lever is located on the right hand side of microscope head

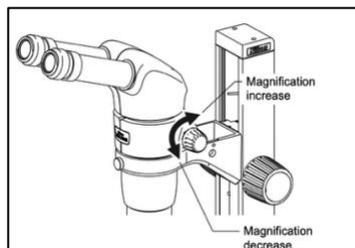
- BINO (100% light to eyes) – Lever fully pulled out
- BINO/PHOTO – Lever fully pushed in (this setting is for image capture)

3. Adjust light intensity
 - a. Ring light control knob located on the power supply at the left of the microscope.
4. Place a sample on the stage
5. Focus on the sample
Utilize the coarse and fine focus knobs to move the zooming body



6. Change the magnification using the zoom knob

There are predetermined stop clicks of magnification on this instrument. Identified on the zoom knob.



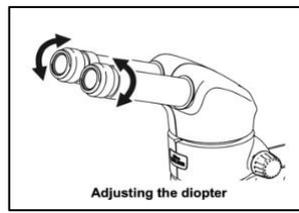
7. Adjust the diopter

The diopter adjustment ring on each eyepiece can be adjusted to match the vision in each eye.

Note: To avoid using your left eye it is a good idea to cover it with a piece of paper. Do not press on your eye as this changes the shape of your lens!

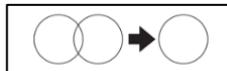
- Focus on your specimen by turning the right Diopter adjustment ring.
- Look into the left eyepiece with your left eye (cover your right eye with a piece of paper). Focus on the sample by turning the left Diopter adjustment ring.
- Repeat the above steps until focus is perfect!

Note: Diopter adjustment increases ease of binocular vision reducing eyestrain and improving imaging. The diopter adjustment ring should always be returned to the diopter adjustment reference position.



8. Adjust the interpupillary distance

When you have the specimen in focus, get the best image possible by looking into the eyepieces using both eyes. Adjust the binocular head such that the distance between your eyes allows the field of view from the left eye and the right eye to coincide. It helps to pretend you are looking into the distance.



Dark-field/OCC Illumination Adjustments

It is typically easiest to setup the microscope for the best images by focusing on the specimen first in bright-field and then transitioning to another technique.

If viewing an opaque/difficult to view specimen, it is recommended to get the microscope setup for viewing in bright-field to your eye strength and then transition to reduce eyestrain/fatigue.

9. Following setup in Bright-field – power off reflected illumination via external power source rocker switch
10. Power on transmitted light power switch via the front of microscope stage rocker switch
11. Adjust illumination via the stage front located illumination knob

Note if no light is transmitted, verify that the OCC Illumination slider is not fully in dark-field.

12. Utilize the slider to obtain the best image.
13. Dark-field is achieved by positioning the OCC illumination slider fully closed.

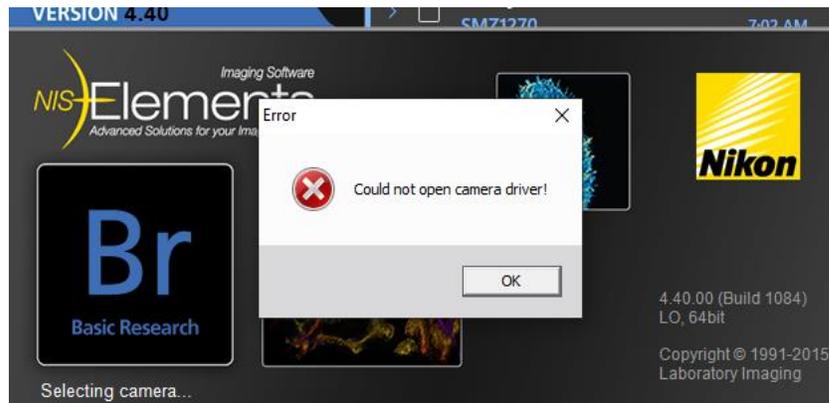
Image Acquisition

1. Turn on the computer
2. Power on the camera

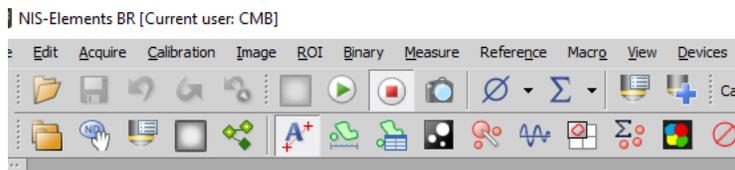
Push button located on camera power supply – to the right of the microscope. Power LED at front will light when powered on.

3. Set the optical path of the binocular tube to distribute 100% light to the camera
 - BINO (100% light to eyes) – Lever fully pulled out
 - BINO/PHOTO – Lever fully pushed in (this setting is for image capture)
4. Open the NIS-Elements microscope imaging software located on the desktop.

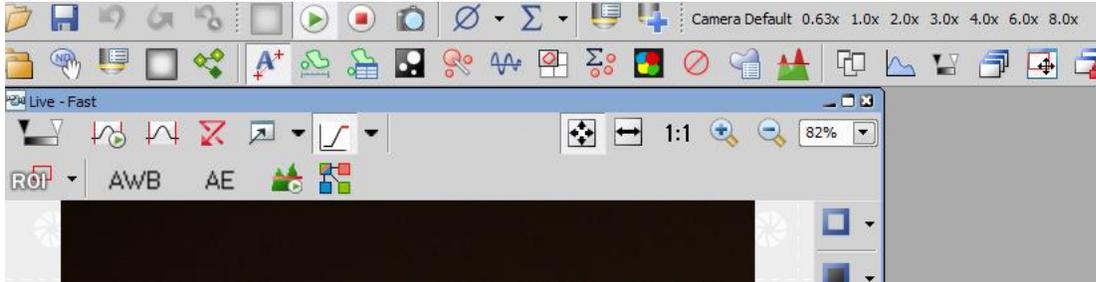
If you open the NIS elements software before camera is fully initialized an error code will pop up (see image below). If that happens, close the error code, exit the NIS elements software and reboot software after camera is powered on.



5. Once software is initialized, to capture an image, you will select the Play  button located in the top tool bar.



6. This will populate an interior window within the NIS Elements software. The top left of that window will state “Live-Fast” to indicate active camera/live feed of your specimen.



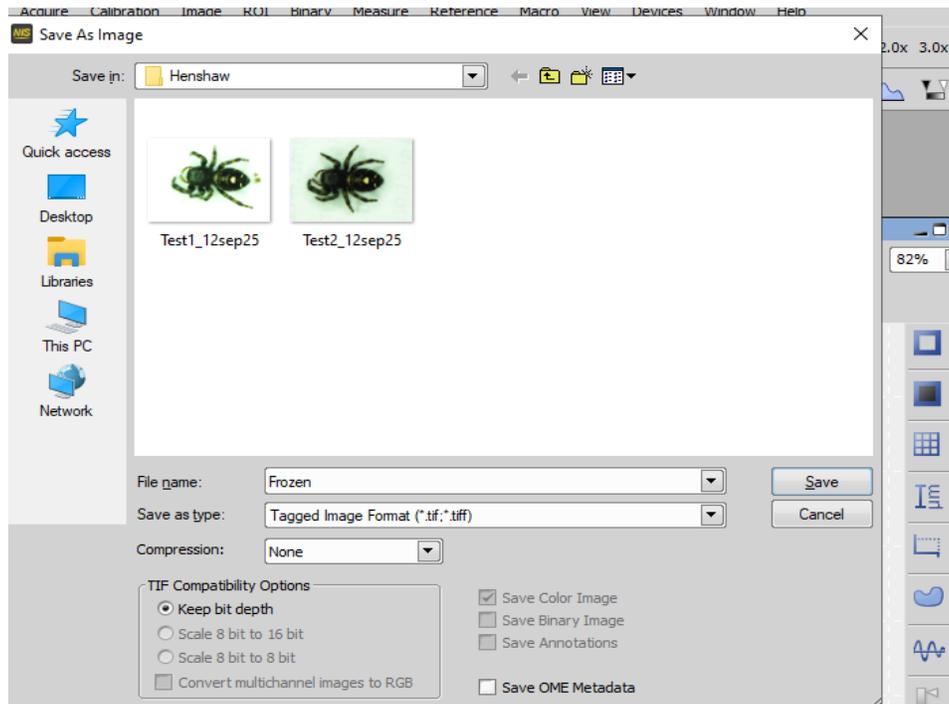
7. To capture an image from “Live-Fast” select the “capture” button . This action will open a second interior window within NIS – Elements (as pictured below). Viewing the top left corner of each window:
 - a. “Frozen” – this is the camera window, the stop icon  is highlighted.
 - b. “Captured” – this is your image.



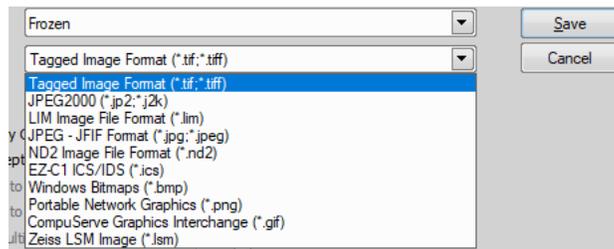
8. To return to your live view, press the play button. The two windows status will be as follows (pictured below):
 - a. "Captured" – image captured
 - b. “Live-fast” – live view of specimen



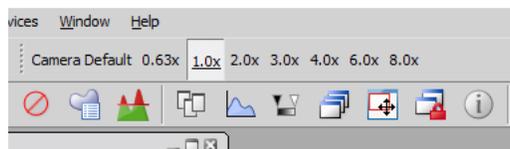
9. To save image: go to File > Save as Image



Pay attention to the file format you are saving your image as. The most popular for after capture image analysis is .Tiff and .Jpeg. If uncertain, ask your PI for their preferred method.



10. Note that there are preprogrammed settings for each magnification selection on the zoom body. It is recommended to verify the settings before image acquisition as these are calibrated to provide accurate measurements/annotations later.



11. Additional Image Analysis information can be found in the NIS-Elements handbook.
There is a copy on each computer with detailed instructions.

Ending your Microscopy Session

It is a courtesy to all to return settings to our “DEFAULT SETTING” to allow the next user to efficiently and easily examine their specimens. It is also setup to prevent any accidental damage to the microscopes, so please follow these instructions.

1. Turn off accessory equipment (camera & computer) if no longer in use
2. Set the optical path switching lever so that 100% of the light is directed to the eyepiece
3. Return the lowest zoom magnification on the zoom body
4. Return the diopter adjustment rings to their reference position
5. Turn off dia-illumination reflective and transmitted sources
 - a. Reflective – rocker switch on external light source box
 - b. Transmitted – rocker switch at the front of the base of the stage of the microscope
6. Cover the microscope

Technical Specifications

- Microscope Make: Nikon
- Microscope Model: SMZ1270
- Eyepieces: CW10xB/22 (with zero reference)
- Microscope Head:
 - 180-degree eyepiece rotation with tilt option
 - BINO/[BINO/PHOTO] lever options
- Objective: PLAN APO 1x WF WD:70
- Zoom body magnification options 0.63-8x
 - 0.63
 - 1
 - 2
 - 3
 - 4
 - 6
 - 8
- Camera: DS-Fi3
 - 5.9 megapixel color microscope camera with high speed data readout (2880 x 2048 pixels)
 - Utilizes CMOS sensor for superior color reproduction
- Camera control unit: DS-U3
- Light sources:
 - Ring light (left hand side of microscope on bench)
 - Schott KL 1600 LED
 - Stage light – bulb (controls located on stage of microscope)