



Introduction to the Microscope

- Types of Microscopes
- Care
- Parts
- Focusing

Types of Microscopes

Light Microscope - the models found in most schools, use compound lenses to magnify objects. The lenses bend or refract light to make the object beneath them appear closer.



Common magnifications:
40x, 100x, 400x

*Oil Immersion lenses
can improve quality of
focus and magnification

Stereoscope

This microscope allows for binocular (two eyes) viewing of larger specimens.

Usually magnifies 10x to 20x

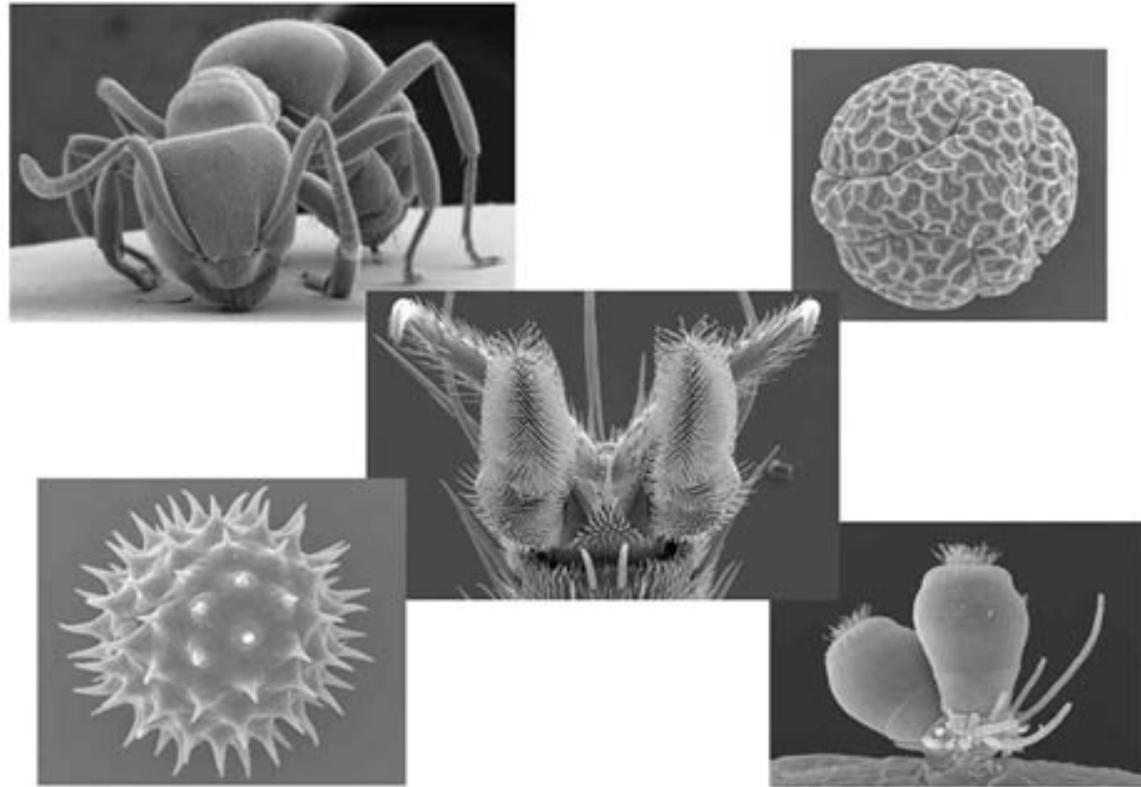
Can be used for thicker specimens

Creates a 3D view of specimen



Scanning Electron Microscope - allows scientists to view a universe too small to be seen with a light microscope. SEMs do not use light waves; they use electrons (negatively charged electrical particles) to magnify objects up to two million times.

SEM creates a 3D view of specimen, but cannot view living specimens (process kills them)

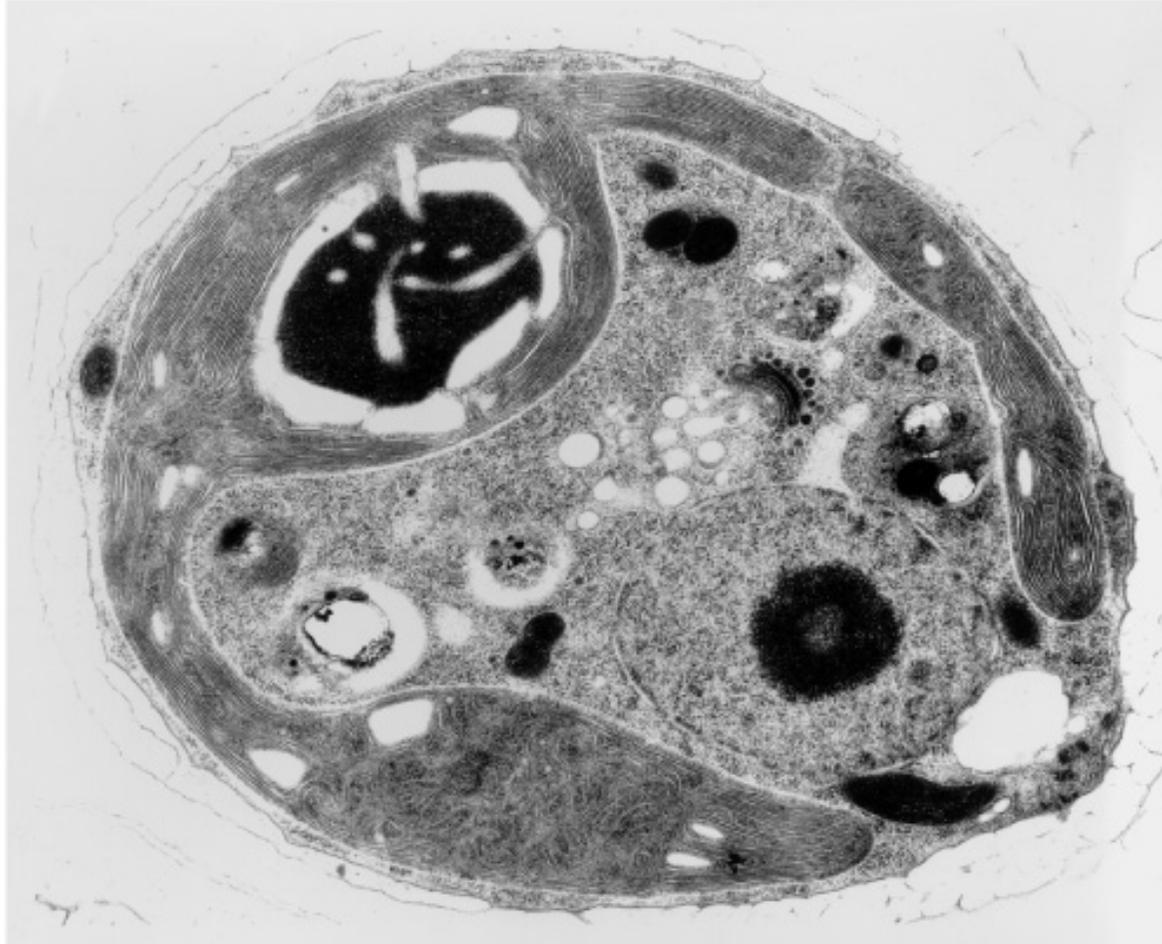


**Transmission
Electron
Microscope** - also
uses electrons, but
instead of scanning
the surface (as with
SEM's) electrons are
passed through very
thin specimens.

TEM = "thin"



TEM of a cell, notice you see the inside of the cell and not the surface.



Microscope Resources

Virtual Microscopes (Phase Contrast, Fluorescence, TEM, STM) at

<http://nobelprize.org/educational/physics/microscopes/1.html>

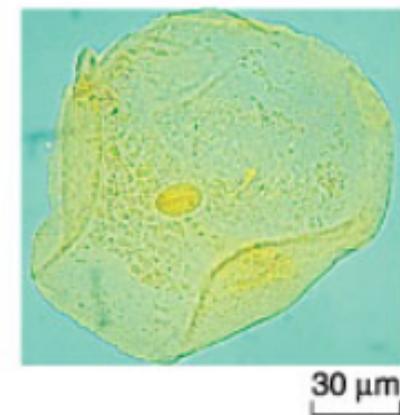
[Nanoworld Image Gallery](#)

[Microworld Images](#)

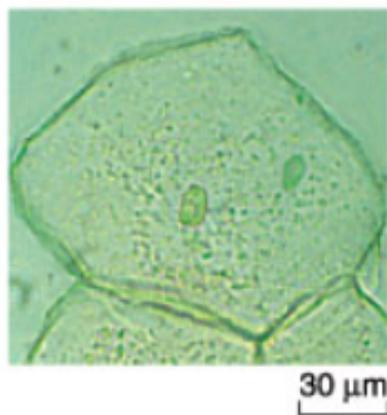
Types of Illumination in Microscopes

How light passes through a specimen changes the view of the specimen, making some parts more distinct.

Copyright © The McGraw-Hill Companies, Inc. Permission required for reproduction or display.



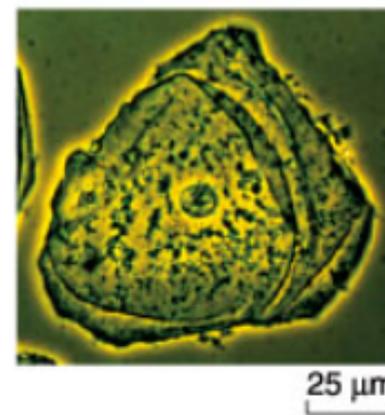
Bright-field. Light passing through the specimen is brought directly into focus. Usually, the low level of contrast within the specimen interferes with viewing all but its largest components.



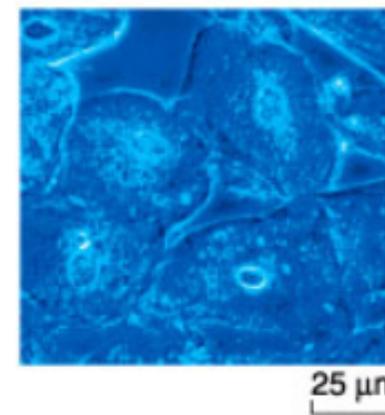
Bright-field (stained). Dyes are used to stain the specimen. Certain components take up the dye more than other components, and therefore contrast is enhanced.



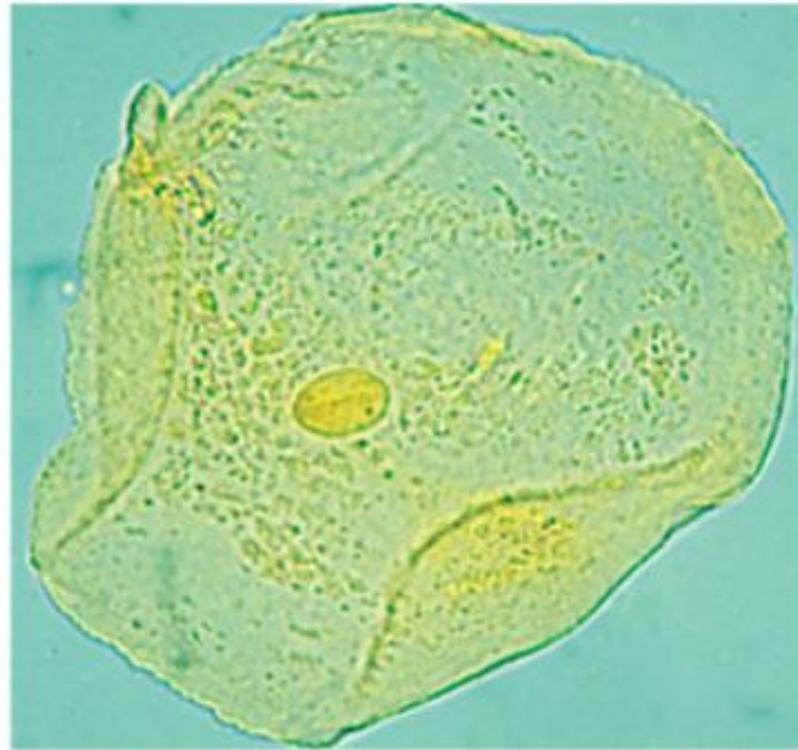
Differential interference contrast. Optical methods are used to enhance density differences within the specimen so that certain regions appear brighter than others. This technique is used to view living cells, chromosomes, and organelle masses.



Phase contrast. Density differences in the specimen cause light rays to come out of "phase." The microscope enhances these phase differences so that some regions of the specimen appear brighter or darker than others. The technique is widely used to observe living cells and organelles.



Dark-field. Light is passed through the specimen at an oblique angle so that the objective lens receives only light diffracted and scattered by the object. This technique is used to view organelles, which appear quite bright against a dark field.



30 μm

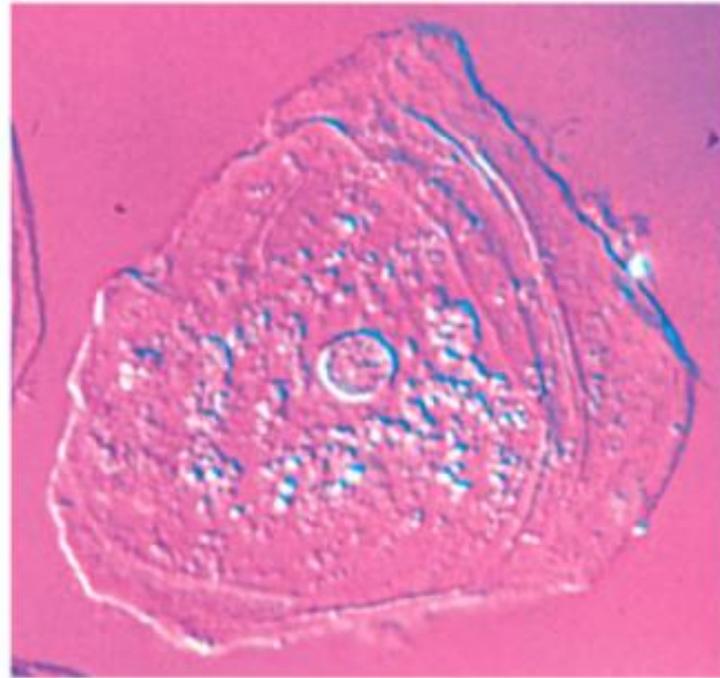
Bright-field. Light passing through the specimen is brought directly into focus. Usually, the low level of contrast within the specimen interferes with viewing all but its largest components.



30 μm

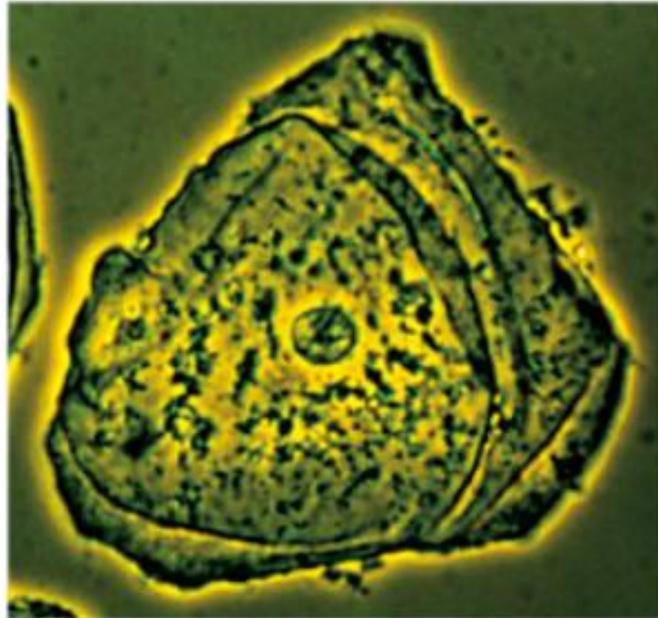
Bright-field (stained).

Dyes are used to stain the specimen. Certain components take up the dye more than other components, and therefore contrast is enhanced.



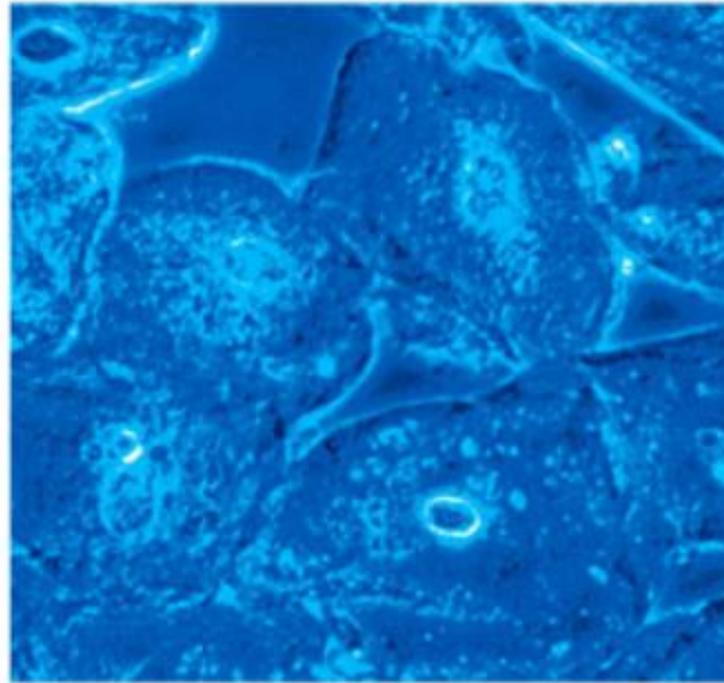
25 μm

Differential interference contrast. Optical methods are used to enhance density differences within the specimen so that certain regions appear brighter than others. This technique is used to view living cells, chromosomes, and organelle masses.



25 μm

Phase contrast. Density differences in the specimen cause light rays to come out of “phase.” The microscope enhances these phase differences so that some regions of the specimen appear brighter or darker than others. The technique is widely used to observe living cells and organelles.



25 μm

Dark-field. Light is passed through the specimen at an oblique angle so that the objective lens receives only light diffracted and scattered by the object. This technique is used to view organelles, which appear quite bright against a dark field.

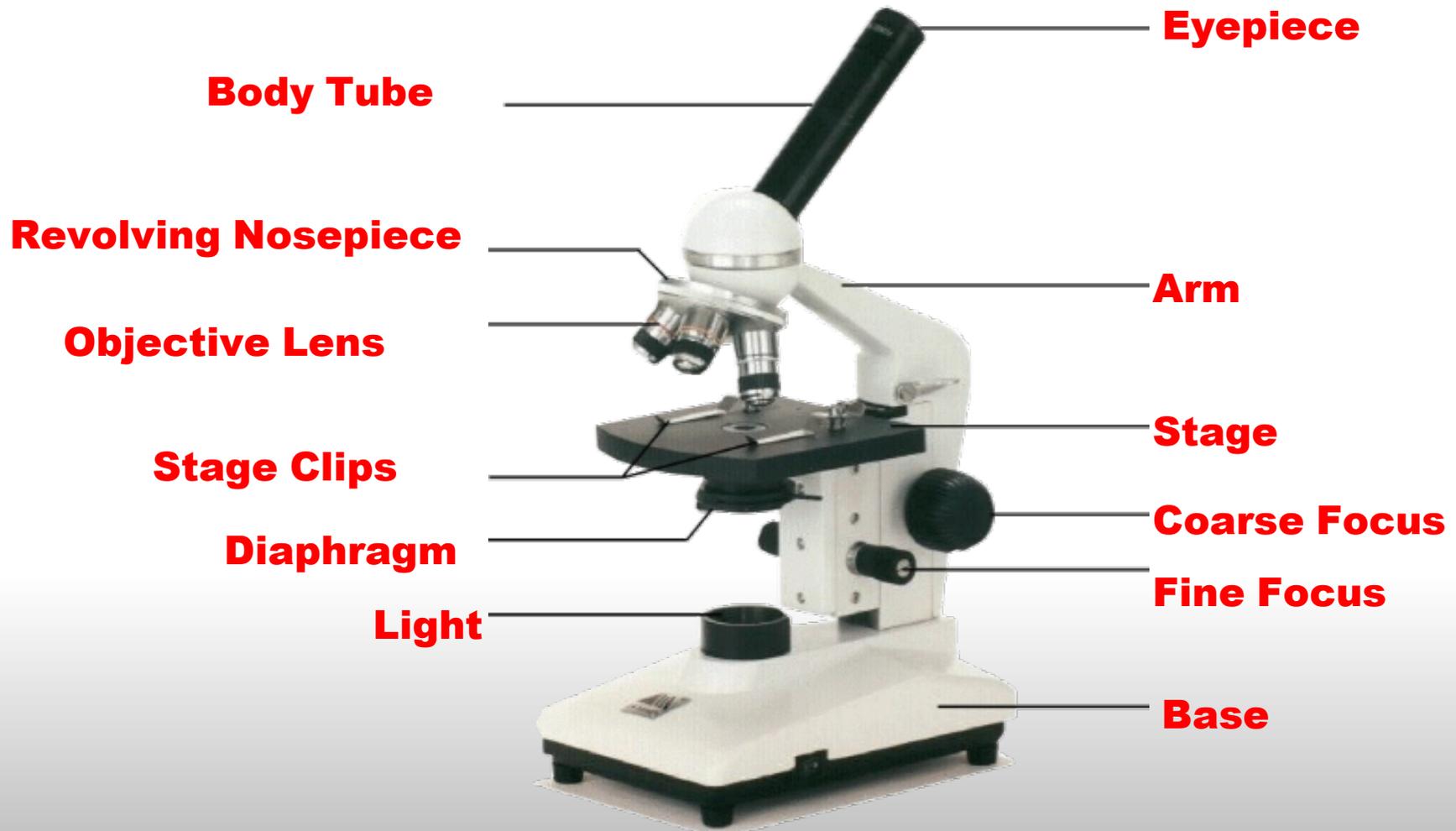
The Light Microscope

Guidelines for Use

- Always carry with 2 hands
- Only use lens paper for cleaning
- Do not force knobs
- Always store covered
- Keep objects clear of desk and cords



Microscope Parts



Magnification

Your microscope has 3 magnifications: Scanning, Low and High. Each objective will have written the magnification. In addition to this, the ocular lens (eyepiece) has a magnification. The total magnification is the ocular x objective

	Magnification	Ocular lens	Total Magnification
Scanning	4x	10x	40x
Low Power	10x	10x	100x
High Power	40x	10x	400x

Using the Microscope

General Procedures

1. Make sure all backpacks and junk are out of the aisles and off the tops of desks.
2. Plug your microscope in to the extension cords. Each row of desks uses the same cord.
3. Store with cord wrapped around microscope and the scanning objective clicked into place.
4. Carry by the base and arm with both hands.

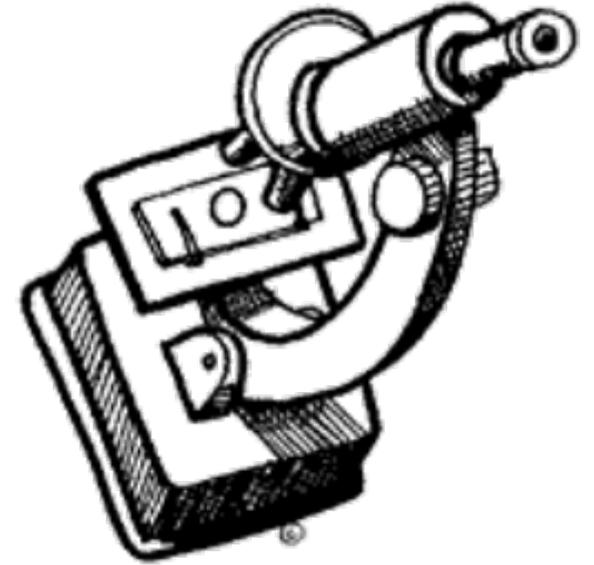


Focusing Specimens

1. Always start with the scanning objective.

Odds are, you will be able to see something on this setting. Use the **Coarse Knob** to focus, image may be small at this magnification, but you won't be able to find it on the higher powers without this first step.

Do not use stage clips, try moving the slide around until you find something.



2. **Once you've focused on Scanning, switch to Low Power.** Use the **Coarse Knob** to refocus. Again, if you haven't focused on this level, you will not be able to move to the next level.

3. **Now switch to High Power.** (If you have a thick slide, or a slide without a cover, do NOT use the high power objective). At this point, ONLY use the **Fine Adjustment Knob** to focus specimens.

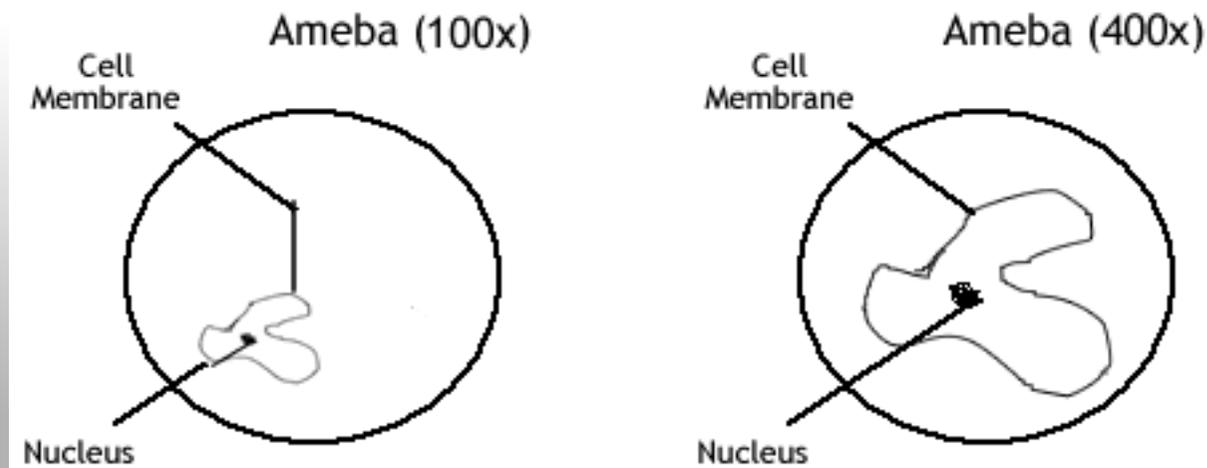
Recap

1. Scanning --> use coarse knob
2. Low power --> use coarse knob
3. High power --> use fine knob

**DO NOT SKIP
STEPS!!!!**

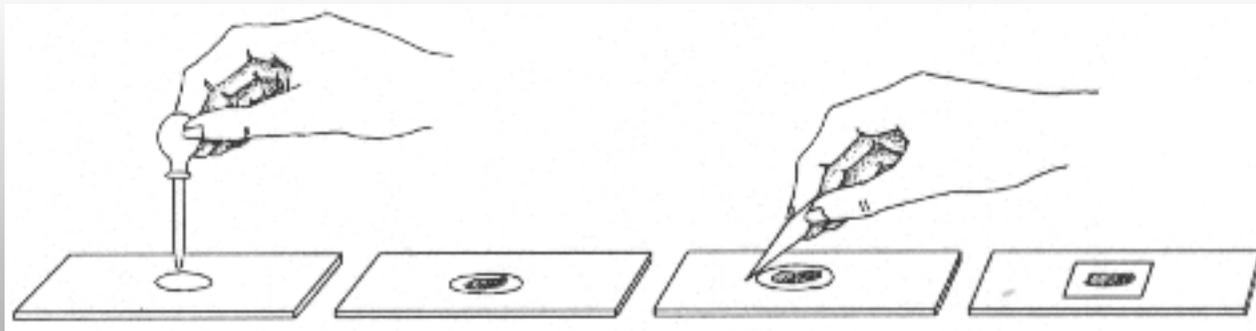
Drawing Specimens

1. Use pencil - you can erase and shade areas
2. All drawings should include clear and proper labels (and be large enough to view details). Drawings should be labeled with the specimen name and magnification.
3. Labels should be written on the outside of the circle. The circle indicates the viewing field as seen through the eyepiece, specimens should be drawn to **scale** - ie..if your specimen takes up the whole viewing field, make sure your drawing reflects that.



Making a Wet Mount

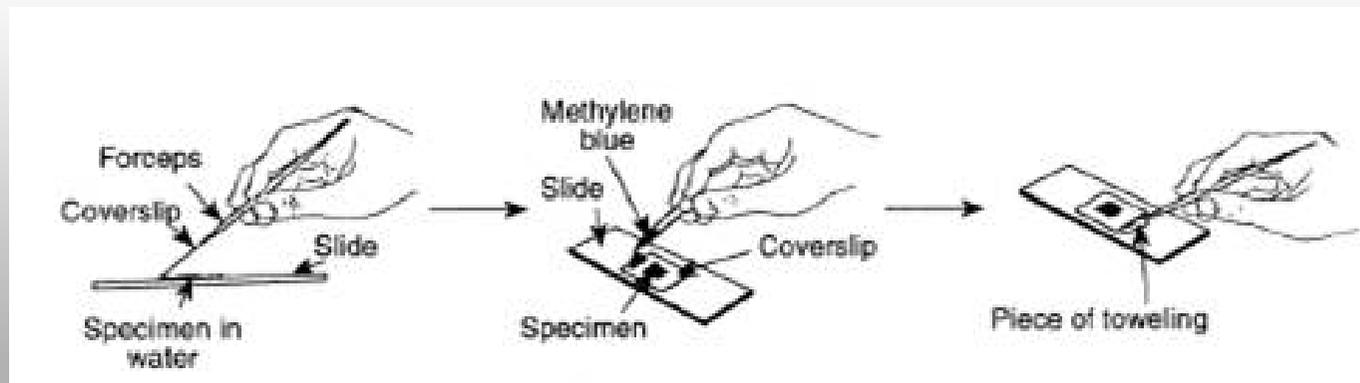
1. Gather a thin slice/peice of whatever your specimen is. If your specimen is too thick, then the coverslip will wobble on top of the sample like a see-saw, and you will not be able to view it under High Power.
2. Place ONE drop of water directly over the specimen. If you put too much water, then the coverslip will float on top of the water, making it hard to draw the specimen, because they might actually float away. (Plus too much water is messy)
3. Place the cover slip at a 45 degree angle (approximately) with one edge touching the water drop and then gently let go. Performed correctly the coverslip will perfectly fall over the specimen.



*Do not drop vertically,
set one edge down and
let the other side drop.*

How to Stain a Slide

1. Place one drop of stain (iodine, methylene blue..there are many kinds) on the edge of the coverslip.
2. Place the flat edge of a piece of paper towel on the opposite side of the coverslip. The paper towel will draw the water out from under the coverslip, and the cohesion of water will draw the stain under the slide.
3. As soon as the stain has covered the area containing the specimen, you are finished. The stain does not need to be under the entire coverslip. If the stain does not cover as needed, get a new piece of paper towel and add more stain until it does.
4. Be sure to wipe off the excess stain with a paper towel.



Cleanup

1. Store microscopes with the scanning objective in place.
2. Wrap cords and cover microscopes.
 - *Double check to make sure you didn't leave a slide
3. Wash slides in the sinks and dry them, placing them back in the slide boxes to be used later.
4. Throw coverslips away. (these are not reusable)
 - *Be careful not to drop these in the sink, they can clog drain.
5. Place microscopes in their designated location (probably a cabinet)

Troubleshooting

Occasionally you may have trouble with working your microscope. Here are some common problems and solutions.

1. Image is too dark!

Adjust the diaphragm, make sure your light is on.

2. There's a spot in my viewing field, even when I move the slide the spot stays in the same place!

Your lens is dirty. Use lens paper, and only lens paper to carefully clean the objective and ocular lens. The ocular lens can be removed to clean the inside. The spot is probably a spec of dust.

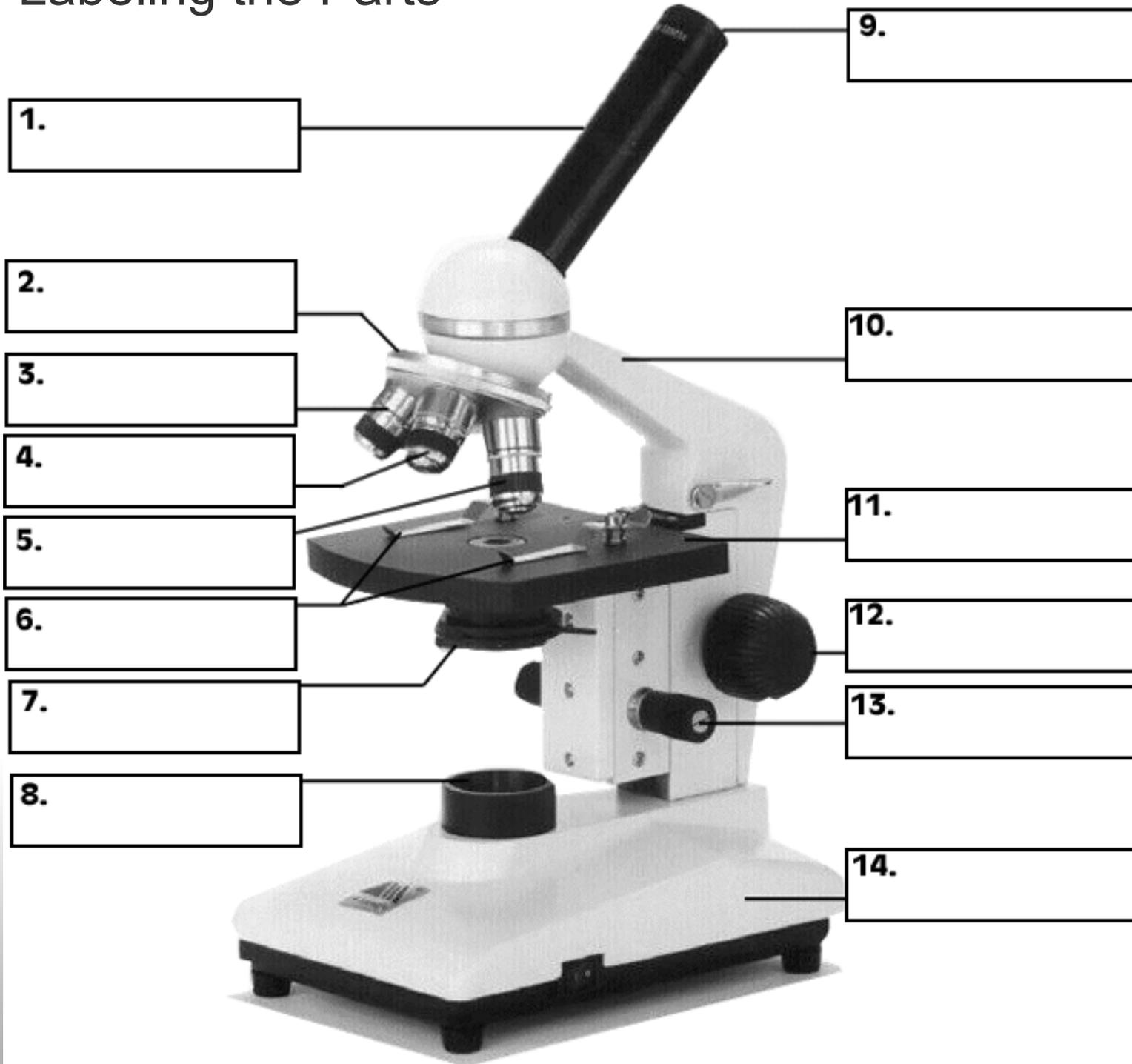
3. I can't see anything under high power!

Remember the steps, if you can't focus under scanning and then low power, you won't be able to focus anything under high power. Start at scanning and walk through the steps again.

4. Only half of my viewing field is lit, it looks like there's a half-moon in there!

You probably don't have your objective fully clicked into place.

Practice Labeling the Parts



Quiz Over the Microscope

1. When focusing a specimen, you should always start with the _____ objective.

2. When using the high power objective, only the _____ knob should be used.

3. The type of microscope used in most science classes is the _____ microscope

4. Stains can be drawn under the slide (and over a specimen) by using a _____

5. What part of the microscope can adjust the amount of light that hits the slide? _____

6. You should carry the microscope by the _____ and the _____.

7. The objectives are attached to what part of the microscope (it can be rotated to click the lenses into place):

8. You should always store you microscope with the _____ objective in place.

9. A microscope has an ocular objective of 10x and a high power objective of 50x. What is this microscope's total magnification? _____

10. SEM is an abbreviation for _____
