

## The Compound Light Microscope

The compound light microscope uses two or more lenses or mirrors to magnify an image. A simple microscope is a single lens (think magnifying glass). The compound light microscope uses visible light as its source of illumination. The source of illumination is the single greatest factor in determining the maximum magnification achievable with the microscope. The greatest magnification possible when using a light microscope is ~1500X. Compound light microscopes can be used to view living or dead specimens. The one characteristic the specimen must possess, is that it must be transparent or translucent. Light must be able to pass through the specimen.

### Important Terms

Magnify: to make larger.

Resolution: the ability to differentiate as separate, objects that are close together.

Field of view: the circle of light seen when the looking through the microscope oculars.

Depth of field: specimens have thickness. As the user focuses up and down through a specimen different layers of the specimen come into focus.

Working distance: the distance between the objective and the stage.

Total magnification: magnification of the specimen calculated by multiplying the magnifying ability of the objective lens by the ocular magnification.

Parfocal: a characteristic of most microscopes. Images in focus at one magnification, will remain in relatively good focus as magnification increases.

### Magnification and Resolution

Magnification means to make something larger. However, just making an image larger is not always revealing. For example, you are driving down the highway and see a yellowish blob on the horizon. You take a picture and then have that image printed as an 8X10 inch glossy photo. The picture reveals a large picture of a blob. The blob has been magnified, it is now much larger but you still don't know what it is. As you drive down the highway and approach the blob, you now see it is a sign for your favorite fast food restaurant. How do you know that? As you get closer, you can resolve the image. You can distinguish any two parts of the sign as separate and unique. Resolution is the important characteristic of the microscope. Resolution is determined by the source of illumination (wavelength) and by the quality of the glass lenses (numerical aperture). When visible light serves as the source of illumination resolution limits the maximum magnification that can be achieved to ~1500X.

### Parts of the Binocular Compound Microscope

## Ocular

The oculars or eyepieces are lenses through which the user looks to see the specimen. The microscope may have one ocular (monocular) or two oculars (binocular). The oculars magnify the specimen. The magnifying ability of the ocular is printed on the ocular, typically the oculars have a 10X magnification. The oculars can be adjusted to accommodate the user's inter-pupillary distance. The oculars should be adjusted so that only a single circle of light is visible when looking into the oculars.

## Head

The head is between the oculars and the nosepiece of the microscope. It contains the mechanism, typically mirrors, to bend the light passing through the stage into the oculars.

## Nosepiece

The nosepiece is attached to the head and holds the objectives. The objectives are rotated over the stage by grabbing and rotating the nosepiece.

## Objectives

The objectives are attached to the nosepiece and are suspended above the stage. The objectives are lenses that aid in magnifying the specimen. The magnifying power of the objective alone is written on the side of the objective. Typical objectives have the following magnifications; 4X (scanning objective), 10X (low power objective), 40X (high dry objective) and 100X (oil immersion objective). The oil immersion lens is used for examining very small organisms, such as bacteria. To use the lens, immersion oil is applied to the slide and the objective is rotated into the oil. If the oil immersion lens is needed, your instructor will provide further instructions.

*The optical elements (objectives, oculars) of your microscope must only be cleaned with lens cleaner and lens paper. Using anything else on the glass elements of the microscope can potentially scratch the glass and destroy the microscope's usefulness.*

## Arm

The arm connects the head to the base. The stage is also attached to the arm. Microscopes should always be carried with two hands. One hand should hold the base, the other holds the microscope's arm.

## Stage

The stage is the platform which holds the slide. The stage has stage clips to hold the slide. Alternatively, the slide is held by the slide clip arm that is part of the mechanical stage.

If anything drips onto the stage from a wet mount the stage should be wiped off with a damp paper towel. Lens cleaner and lens paper do not need to be used on the stage. The stage should be lowered to its lowest position for storage.

## Mechanical stage

The mechanical stage sits on top of the stage itself. It has a clip arm to hold the slide. The mechanical stage is controlled by knobs which are attached to the clip arm and hang down below the stage on one side the stage. One knob moves the mechanical stage from right to left, the other knob moves the mechanical stage from front to back of the stage.

## Condenser

The condenser is located below the stage. It focuses light from the light source, through the stage opening and specimen.

## Iris diaphragm

The iris diaphragm or diaphragm is found below the condenser. A lever on the diaphragm slides right to left. Sliding the lever in one direction opens the diaphragm and allows more light to pass through the specimen. Moving the lever in the other direction decreases the amount of light passing through the specimen.

## Coarse focus adjustment knob

The coarse adjustment knob is the larger outer knob attached to the arm near the base. Turning the coarse adjustment knob leads to the visible up and down movement of the stage. The coarse adjustment knob is used to do the initial focusing of the specimen. Because most microscopes are parfocal, once the specimen is brought into focus with the coarse adjustment knob, it should only require fine focusing when changing to higher magnification.

## Fine focus adjustment knob

Inward from the coarse adjustment knob is a smaller knob, the fine focus adjustment knob. Once the specimen has been focused with the coarse adjustment knob, the fine adjustment knob is rotated to bring the specimen into sharp focus. When the next higher power objective is rotated over the stage the specimen should only require fine adjustment.

## Base

The microscope sits on the base. The base holds the illuminator and light controls. The location of the light controls depends on the microscope brand; controls are commonly located on the arm or base.

## Illuminator, sub-stage light

The illuminator is the light source.

## Light control/rheostat

The light control/rheostat regulates the amount of light produced by the illuminator. The light control/rheostat may be located on the base or on the arm.

## Power switch

The power switch turns on the light. The power switch may be located on the base or on the arm of the microscope.