University Terms & Definitions

Achromatic Lens (without color)

A lens consisting of two or more elements usually made of crown and flint glass. This lens has been corrected for chromatic aberration in that it has the same focal length with respect to two selected colors (red and blue) or wavelengths of light. The resultant image is free of extraneous colors. It is also corrected for spherical aberration with respect to one color.

Chromatic Aberration

An optical defect of a lens which causes different colors or wave lengths of light to be focussed at different distances from the lens. It is seen as color fringes or halos along edges, and also degrades the image quality.

Compound Microscope

An exacting optical instrument used to magnify and resolve fine detail within a transparent specimen. It differs from the simple microscope (ordinary magnifier) in that it has two separate lens systems; an objective, located near the specimen, which magnifies by the specimen a definite amount, and an eyepiece which further magnifies the image formed by the objective. The resultant magnification observed by the eye is equal to the product of the primary magnification of both lens systems.

Condenser

The lens or lens system to collect illumination light rays and converge them to a focus. Located directly beneath the microscope stage. Coupled with aperture diaphragm, the condenser constitutes one of the most important and necessary features of a good microscope. For this reason, microscopes are equipped with condensers and diaphragms to controllably increase resolution, enhance contrast, reduce glare and assure optimum results with all objective-eyepiece combinations.

Cover-Glasses (coverslips)

Square, rectangular or circular coverslips of thin, optically flat glass used to cover microscope slide specimens. The thickness of the cover glass affects the light rays and most microscope manufacturers design objectives for use with cover glasses having a thickness of 0.17mm. It is particularly recommended that cover glasses with a thickness of 0.17± .2mm be applied to all specimen slides which will be observed critically through an objective of 40x and higher magnification.

Critical illumination

In critical illumination, the image of the light source is focused onto the specimen to create bright illumination. Critical illumination utilizes pre-aligned condenser, making it the perfect choice for students and beginners.

Field Diaphragm

A diaphragm limiting the field of view. It should be adjusted for each objective so that the leaves are just outside the field of view.

Field of View

The visible area seen through the microscope when a specimen is in focus. It is usually expressed in mm diameter when a specimen is in focus. It can be determined by focusing onto a finely graduated, transparent millimeter scale placed on the microscope stage. The field of view varies inversely, with the resultant magnifications- the greater the magnification, the smaller the field of view.

Filter

A transparent material characterized by selective absorption of light according to wavelength.

Flatness of Field

Appearance of the image to be flat, a plane in the object is imaged as a plane.

Iris Diaphragm

An assembly of thin metal leaves controllable by a lever to produce variable sized opening. It is generally associated with microscope condensers and illuminators of intermediate and advanced types.

Kohler Illumination

Kohler illumination is an illumination technique that provides optimum resolution and contrast in a light microscope by aligning and focusing the illumination, and critically setting the apertures of the microscope to best match the objective lens Numerical Aperture.

Microscope

A high precision optical instrument which uses light to study the fine detail of objects. It is capable of high magnification and is used for making minute details visible.

- Bright field microscope- the most ordinarily used in laboratory work. Stained slides are usually used
- Dark field microscope- makes the specimen appear luminous against a background of little or no light. Used for objects displaying feeble contrast in a bright field microscope. Incredibly useful for transparent or low contrast specimens or cell details that would otherwise be invisible under brightfield.
- Metallurgical microscope- designed for visual examination, with magnification of opaque objects, polished metal specimens, and similar materials.
- Phase contrast microscope- used for viewing living specimens or other low contrast specimens which would ordinarily not be visible in the bright field microscope. This microscope uses the principles of diffraction also refraction and scattering. Interference is also a factor to show up the slight differences in optical path
- Polarizing microscope- employs polarized light to show changes in internal structure and composition of material not discernible with ordinary light. Used extensively in industry for product control. Ideal method for viewing crystal and its structures.
- Stereoscopic wide field upright microscope- used to give a three dimensional view of a large specimen. Has a limited magnification range (to about 2000x)

Numerical Aperture (N.A.)

A designation, usually engraved on objectives and condensers, expressing mathematically the solid cone of light delivered to the specimen by the condenser and gathered by the objective. The higher the numerical aperture of an objective the greater the resolving power of the objective. To satisfy this condition, however, it is necessary that the N.A. of the condenser be equal to or greater than the N.A. of the objective. For example, a condenser with N.A. 0.55 is insufficient to drive the full resolving power of a 100x oil immersion objective rated at N.A. 1.25.

Objective

The complex lens system located directly above the object or specimen. It is the most exacting sub-assembly of a microscope since it is called upon to faithfully magnify and resolve specimen detail.

Objectives are used in determining the magnification and resolution for observation in the microscope:

- Achromatic objectives have a hemispherical front lens and a meniscus second lens which work together to bring two different wavelengths to a common focus with a minimum of spherical aberration
- According to organization of standardization (ISO), there are three groups of objectives differentiating in the quality of chromatic, spherical and flatness of field.
- Focusing errors can be caused by field curvature (lens is curved, sharpness of the image maybe a bit off at the outer edge of field of view
- To correct for focusing error, most manufacturers are starting to provide flat-field correction for achromat and plan achromat objectives

Limits of Magnification

Theoretically, there are no upper limits to magnification. However, when magnification exceeds the limits of resolution, the image drops out of focus and features become less defined. This phenomenon is called empty magnification, where resolution is unable to match the magnification.

Useful magnification, or the range of magnification in which magnification does not exceed resolution is largely determined by the microscope's objective lenses. Numerical aperture or N.A. for short is the unit that determines the cone size of light that the objective can accept. The higher the numerical aperture, the higher the resolution that is achievable with your microscope. The best magnification range is usually 500-1000 times the numerical aperture.

Types of Objectives:

Objective Class	Field Flatness	Absolute Value(s) of Focus	Application
Achromats	70%	Between the red and blue wavelengths. Equal or less than 2X depth of field	For standard applications in the visual spectral range
E-Plan Achromats	85%	For red, blue and green wavelengths. Equal or less than 2.5X depth of field	For applications in the visual spectral range with higher specifications
Plan A Achromats	95%-100%	For red, blue and green wavelengths. Equal or less than 1X depth of field	For applications with highest specifications in the visual range and beyond

Parcentered:

A tern applied to the objective indicating that when a specimen detail is in the center of the field of view of one objective, the detail will essentially remain in the center of the field when the next objective is rotated into position.

Parfocal

A term applied to objectives and eyepieces when practically no change in the focus has to be made when on power is substituted for another. The objectives on the revolving nosepiece of a microscope are parfocalized so that only a slight turn of the fine adjustment is required when changing from a low to a higher power objective.

Phase Contrast

A special method of controlled illumination, for observing thin transparent objects whose structural details vary only slightly in thickness and refractive index and, therefore, are not visible in the bright field microscope. This method involves interference of a portion of the light with the rest in such a manner as to produce a visible image.

Polarized Light

Which is vibrating in one plane only. Normal emitted light (a mixture of light waves vibrating in all directions) may be polarized by reflection, double refraction, selective absorption, or scattering. Polarization enables distinguishing the changes in structure and the composition of material that are not discernible with ordinary light. Change in appearance under the polarized light serves as identification.

Units of Measurement

1 meter (m) = 100 mm 1 millimeter (mm)= .001m

1 micron (mu) = .001 mm 1 millmicron (m mu)= .001mu = .001mu = .001.001mm 1 Angstrom (A) = .1m mu =.001,000 1mm

Working Distance

The distance between the front mount of the objective and the top of the cover glass when the microscope is focused on a thin specimen preparation. The greater the primary or initial magnification of the objective, the smaller the working distance.