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CULTURING AND VISUALIZING CELLS

REVIEW THE CONCEPTS

1. Electron microscopy has better resolution than light microscopy, but many light-microscopy techniques allow observation and manipulation of living cells.

2. The total magnification of an image is described as the product of the magnification of the individual lenses, where the objective lens magnification immediately above the specimen is multiplied to that of the projection or eyepiece lens. Being able to clearly distinguish between two closely spaced points at even the highest total magnification is the ultimate goal because if the two objects are already blurred and cannot be discriminated at a lower magnification, simply increasing the magnification will have no effect. In fact, the formula defining the resolution (D) of a lens does not take magnification into account and is written as D = 0.611 N sin a, where 1 is the wavelength of light used to illuminate the specimen, N is the refractive index of the medium (usually air) between the front face of the objective lens and the specimen, and a is the half-angle of the cone of light entering the face of the objective lens. N sin a is often referred to as the lens' numerical aperture, which is physically stamped on the barrel of the objective lens. Since only three of the values can be altered to achieve the best resolution (the smallest D possible), one has to either decrease the wavelength of light or increase the numerical aperture by gathering more light into the front face of the objective lens. In most circumstances, therefore, the limitations include the use of wavelengths in the visible spectrum and the ability to gather more light to increase the numerical aperture. Increasing the numerical aperture is accomplished by placing a drop of oil or water, which have greater refractive indices (1.5 and 1.3, respectively) relative to that of air (1), between the specimen and the objective lens.