Optics primer

Cleaning optics

Geometric optics

Aberrations

Fourier optics

Modern microscopy in Aph162 lab

Cleaning optics

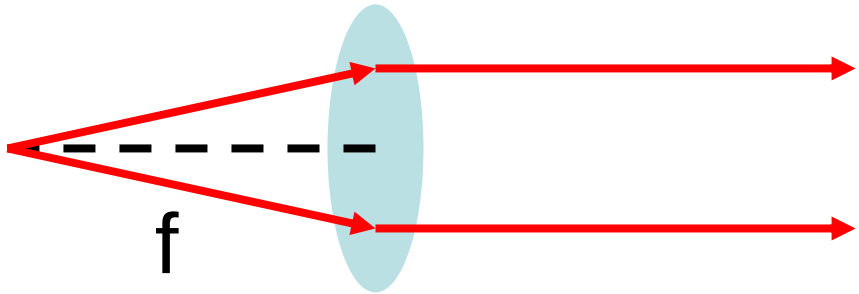
- Don’t clean optics… but if its dirty

Bare optics:
1)Blow off dust
(not with organics)
2)Drag and drop
* Make sure solvent
is good for coatings
* don’t clean bare metal
surfaces with tissues
* Don’t contaminate lens tissue

Cleaning optics

- Objectives
  - 1) roll up lens tissue
  - 2) blot off excess oil
  - 3) roll up lens tissue and apply solvent; shake off excess
  - 4) wipe center outward and discard.

Geometric optics



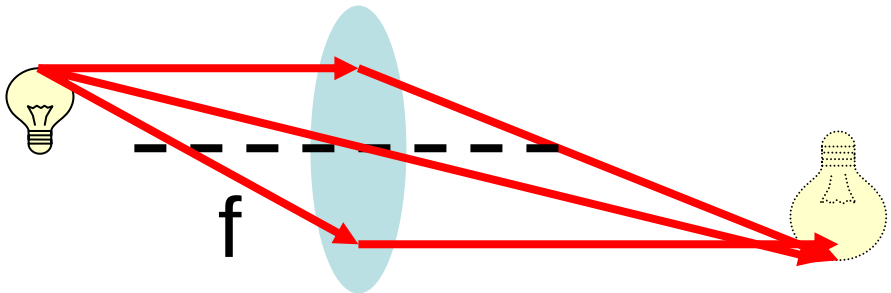


Image is real

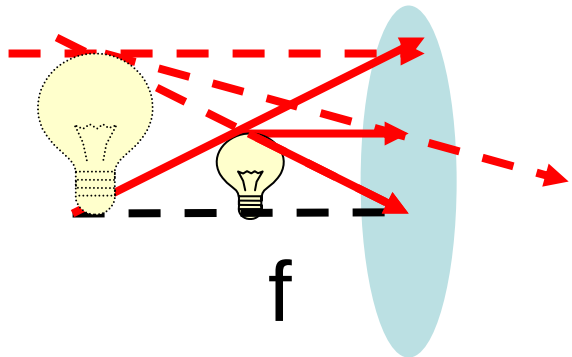
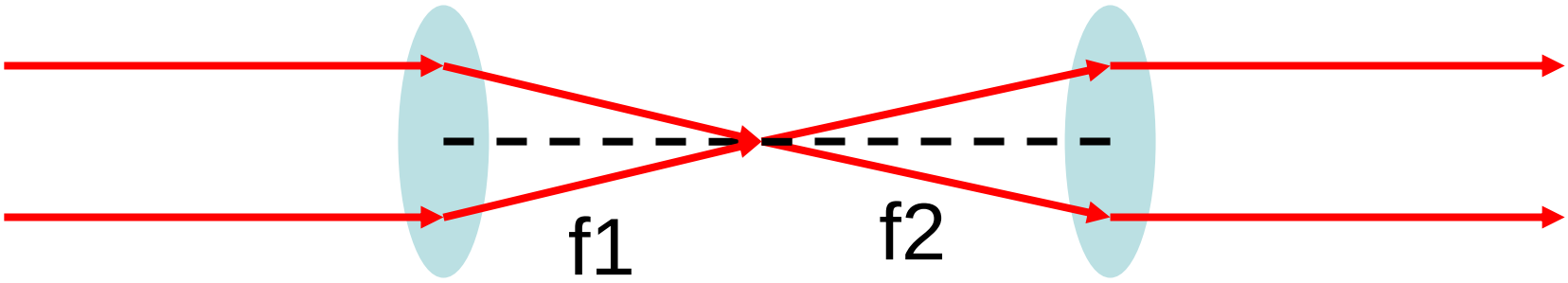


Image is virtual

Magnification

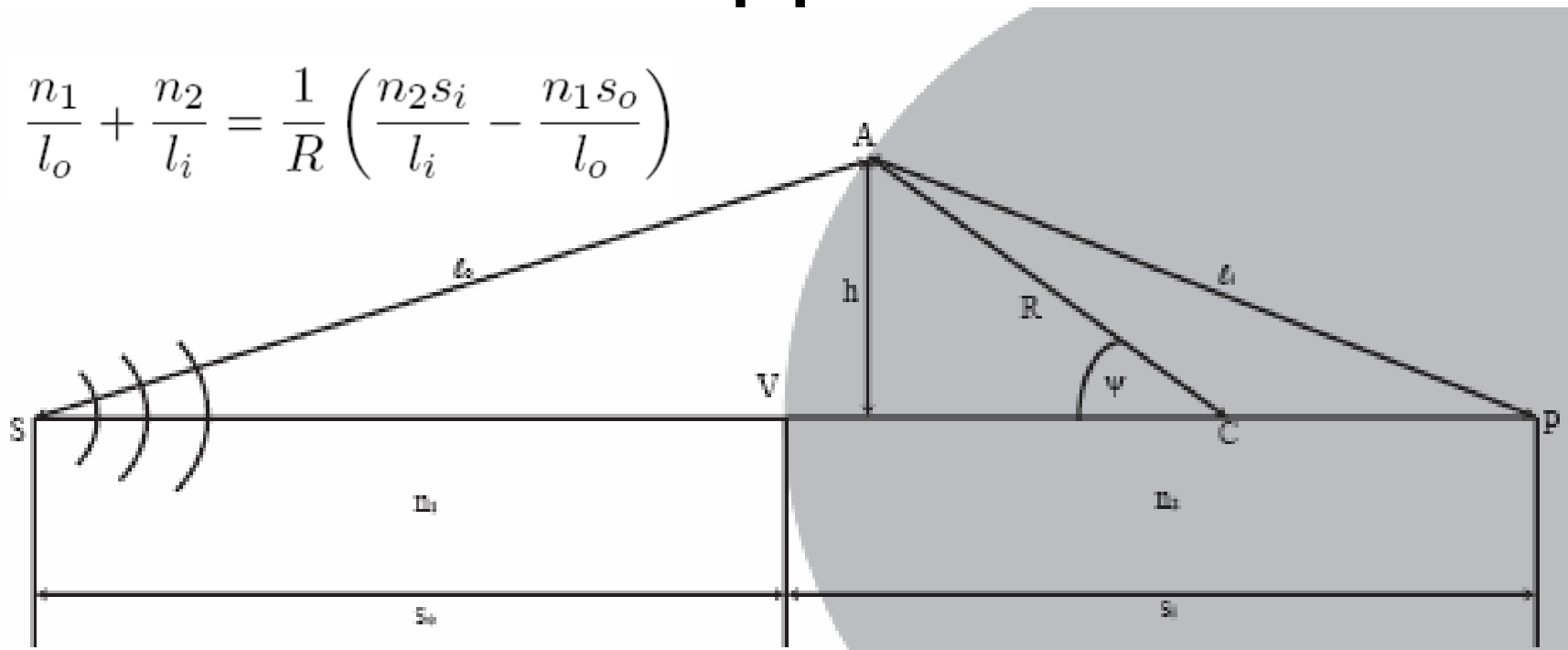


M = f1/f2

M = f1/f2

Paraxial approximation

$$\frac{n_1}{l_o} + \frac{n_2}{l_i} = \frac{1}{R}\left(\frac{n_2 s_i}{l_i} - \frac{n_1 s_o}{l_o}\right)$$

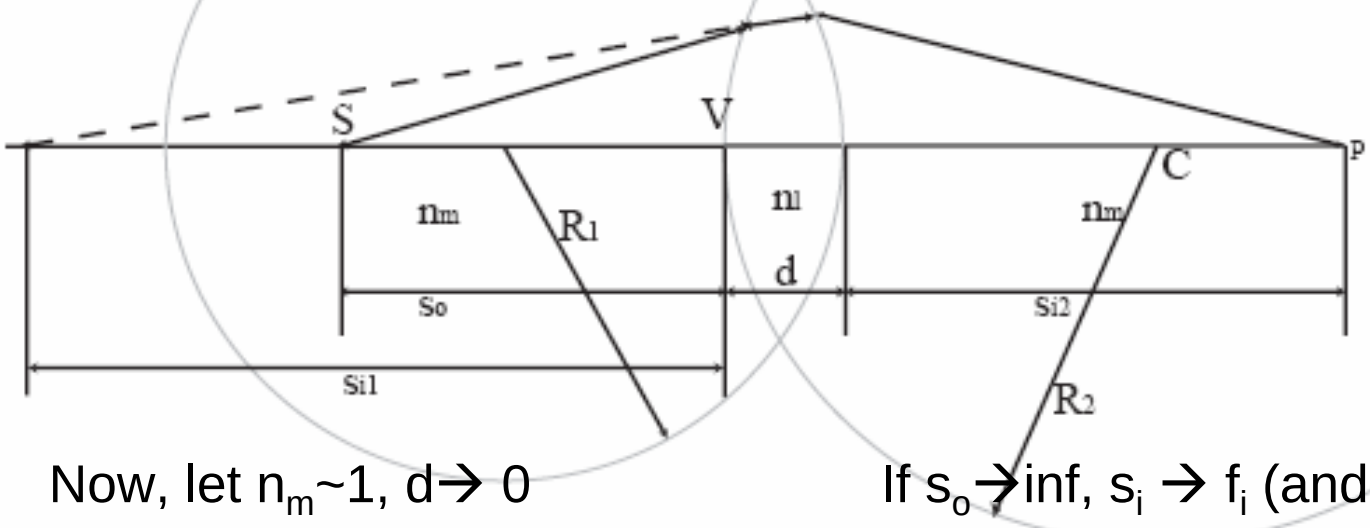


For small ψ, $cos\psi \approx \psi$ and $sin\psi \approx \psi$ so $l_o \approx s_o$ and $l_i \approx s_i$

$$\longrightarrow \quad \frac{n_1}{s_o} + \frac{n_2}{s_i} = \frac{1}{R}(n_2 - n_1)$$

Thin lens equation

$$\frac{n_m}{s_o 1} + \frac{n_m}{s_i 2} = \left(\frac{1}{R_1} - \frac{1}{R_2}\right)(n_l - n_m) + \frac{n_l d}{s_i 1\,(s_i 1 - d)}$$



Now, let $n_m \sim 1$, $d \to 0$

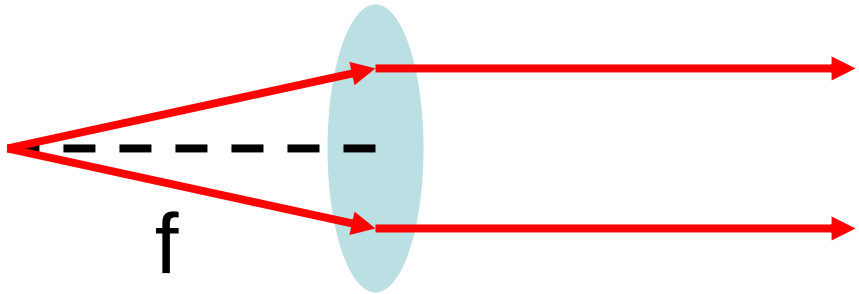
$$\frac{1}{s_o} + \frac{1}{s_i} = (n_l - 1)\left(\frac{1}{R_1} - \frac{1}{R_2}\right)$$

If $s_o \to \infty$, $s_i \to f_i$ (and vice versa)

$$\frac{1}{s_o} + \frac{1}{s_i} = \frac{1}{f}$$

Gaussian lens formula

Geometric optics



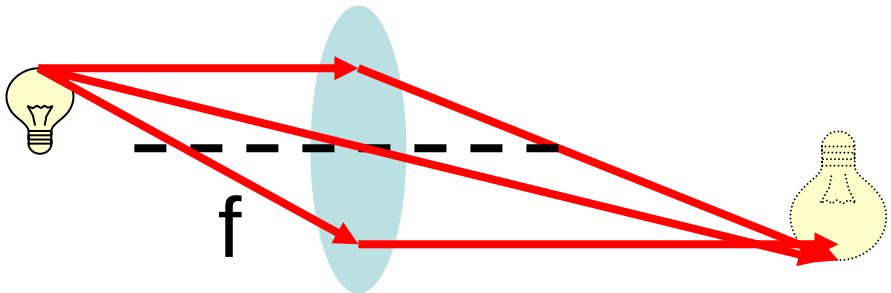


Image is real

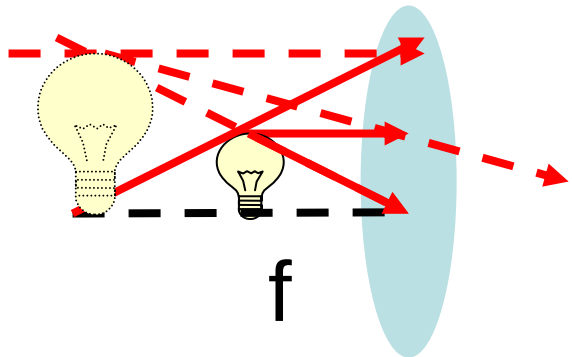
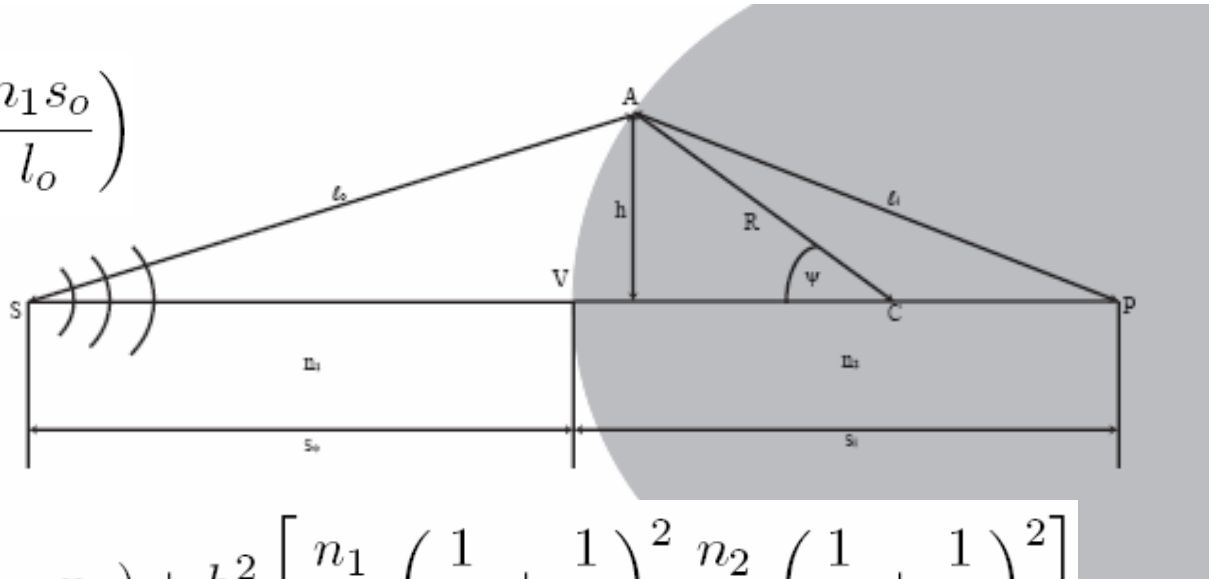


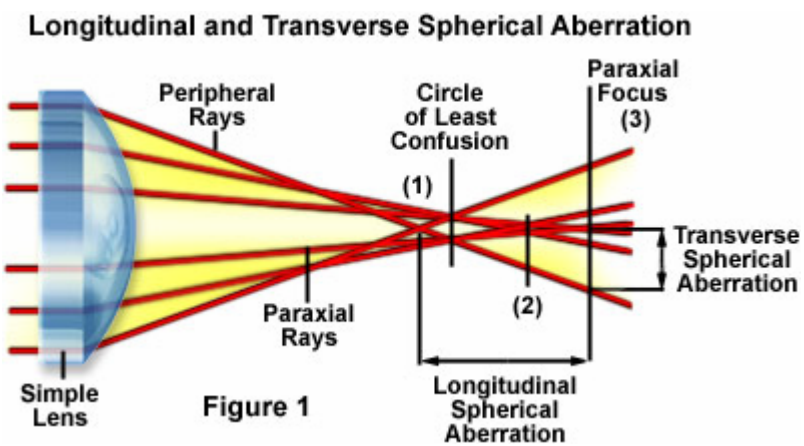
Image is virtual

Higher approximation (primary abberrations)

$$\frac{n_1}{l_o} + \frac{n_2}{l_i} = \frac{1}{R}\left(\frac{n_2 s_i}{l_i} - \frac{n_1 s_o}{l_o}\right)$$



$$\frac{n_1}{s_o} + \frac{n_2}{s_i} = \frac{1}{R}(n_2 - n_1) + h^2\left[\frac{n_1}{2s_o}\left(\frac{1}{s_o} + \frac{1}{R}\right)^2 \frac{n_2}{2s_i}\left(\frac{1}{s_i} + \frac{1}{R}\right)^2\right]$$

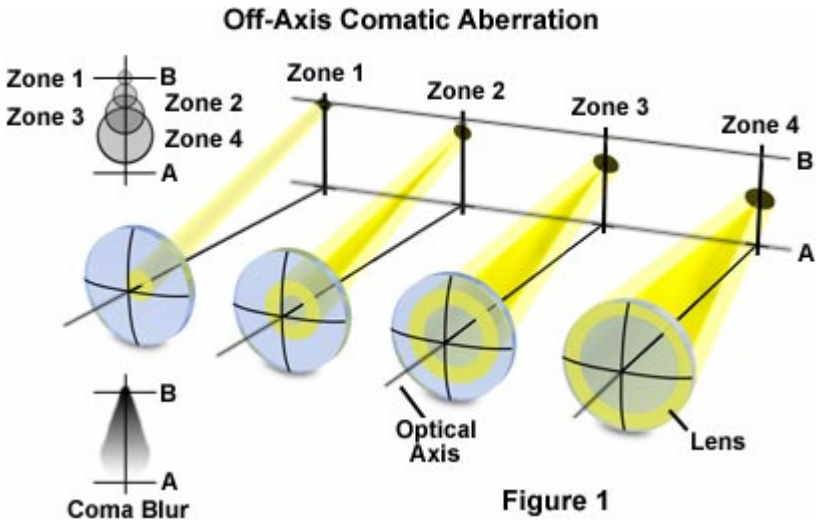


Fixes:
>Apertures
>1 surface (PCX→BCX)
>aspherics
>doublets

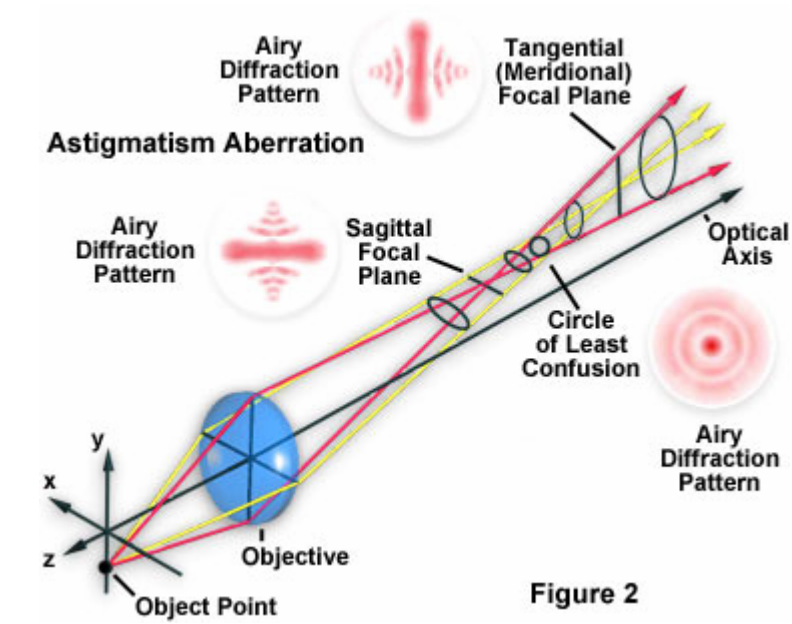
Actually, this lens is backwards…

Aberrations

- Coma



- astigmatism



Aberrations

- Field curvature

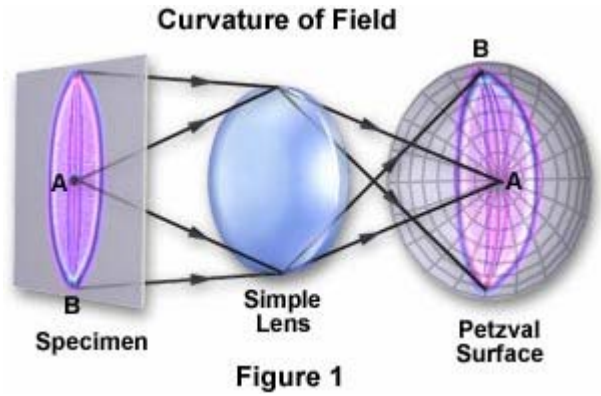
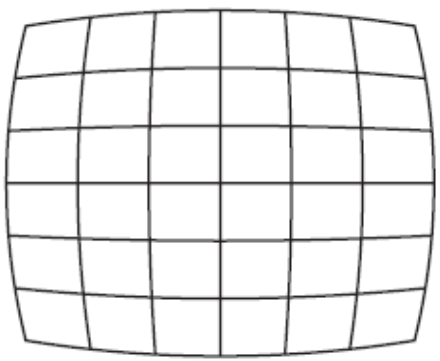
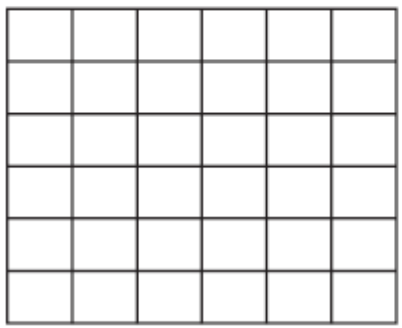
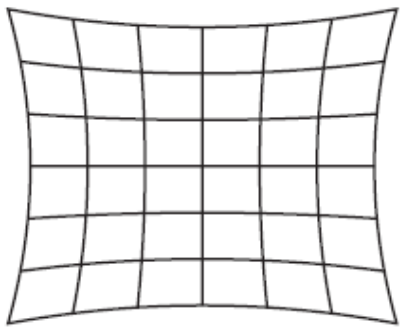


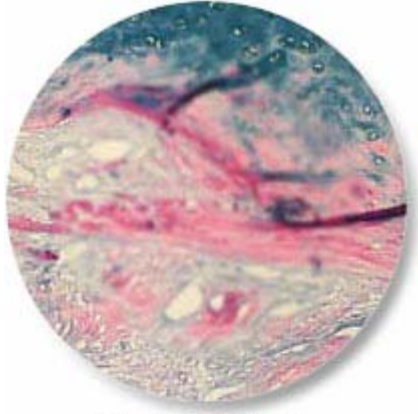
Figure 1

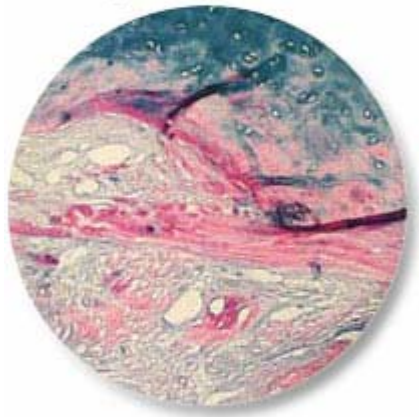
- Distortions








(a) Edges in Focus


(b) Entire Viewfield in Focus

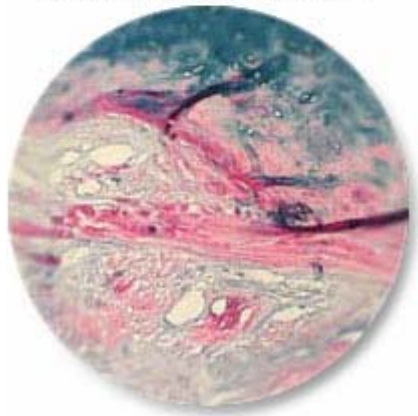

(c) Center in Focus

Figure 2

Aberrations

- Chromatic aberration

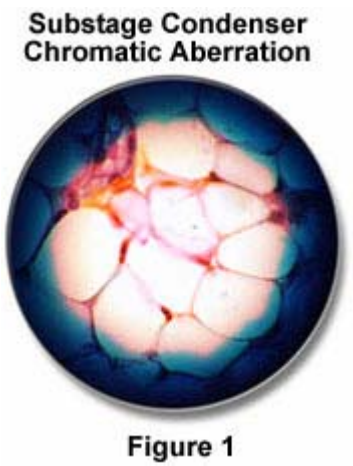


Figure 1

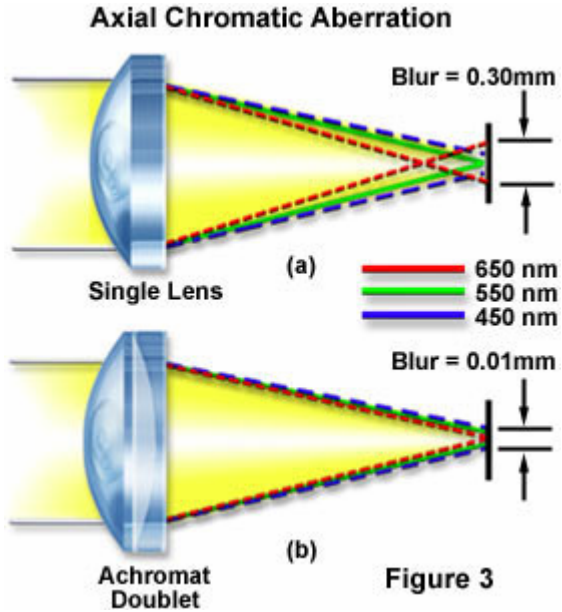


Figure 3

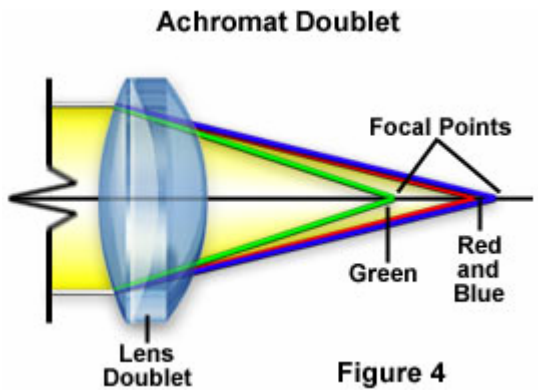


Figure 4

Fourier Optics

$$u(r,t) = \mathrm{Re}[A(r)\exp(-i2\pi\nu t + i\phi(r))]$$

satisfies

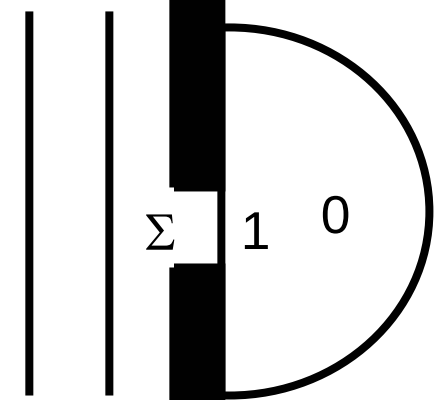
$$\nabla^2 u - \frac{n^2}{c^2}\frac{\partial^2 u}{\partial t^2} = 0$$

$$\downarrow$$

$$(\nabla^2 + k^2)\underbrace{A(r)\exp(i\phi(r))}_{U(r)} = 0$$

$$k = \frac{2\pi}{\lambda} = \frac{2\pi n\nu}{c}$$

Fourier Optics



1) Outgoing waves only (Sommerfeld radiation condition)
2) Field through opening not influenced by aperture
3) Over opaque screen, field is zero
4) →Aperture dimensions >> λ (neglect fringing)
5) Aperture to observation is far compared to wavelength

$$U(0) \propto \iint_{\Sigma} U(1) \frac{e^{ikr_{01}}}{r_{01}} \cos\theta \, ds$$ (Huygens-Fresnel principle)

θ is angle between observation and normal to aperture

$$U(x,y) \propto \iint U(\xi,\eta) e^{\frac{ik}{2z}(\xi^2+\eta^2)} e^{-\frac{i2\pi}{z\lambda}(x\xi+y\eta)} d\xi d\eta$$

(Fresnel Approximation)

Good to distances very close to aperture

Fourier Optics

$$z >> \frac{k\left(\xi^2 + \eta^2\right)}{2}$$ Fraunhofer Approximation

$$\downarrow$$

$$U(x,y) \propto \iint U(\xi,\eta) e^{-\frac{i2\pi}{z\lambda}(x\xi + y\eta)} d\xi d\eta$$

For visible light, and 1 inch aperture, z is 1600 meters!

Note: spherical waves passing through an aperture is diffraction.

FT of a circular aperture is an Airy pattern – how we define optical resolution – width of central lobe

$$d = 1.22\frac{\lambda z}{w} \longrightarrow \frac{\lambda}{2NA}$$ Rayleigh criterion

Fourier Optics

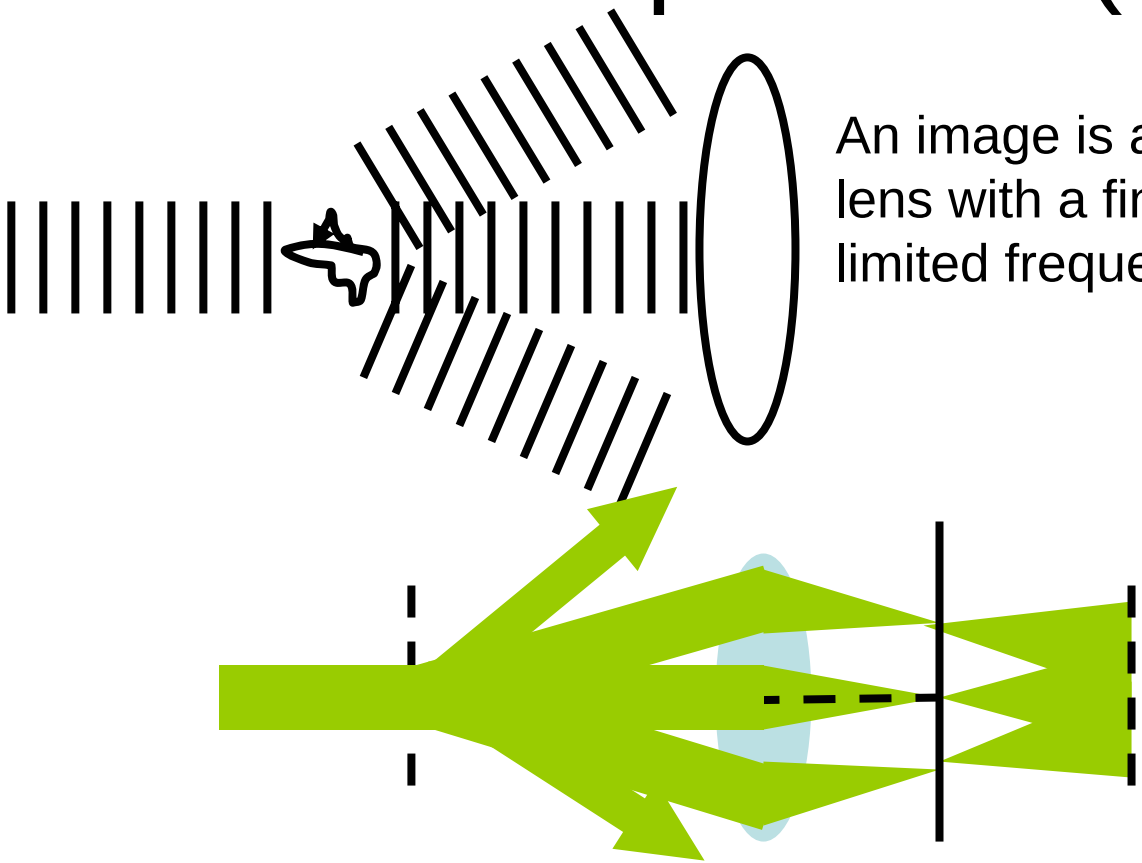
So, light diffracts off an object – and we collect it with a lens. What's going on? – The lens moves the far-field diffraction pattern closer.

Amplitude function behind a lens is:

$$U(x,y) = \iint T(\xi,\eta) e^{-\frac{i2\pi}{f\lambda}(x\xi+y\eta)} d\xi d\eta$$

Thus, a lens computes the Fraunhofer diffraction pattern.

Consequences (E. Abbe)



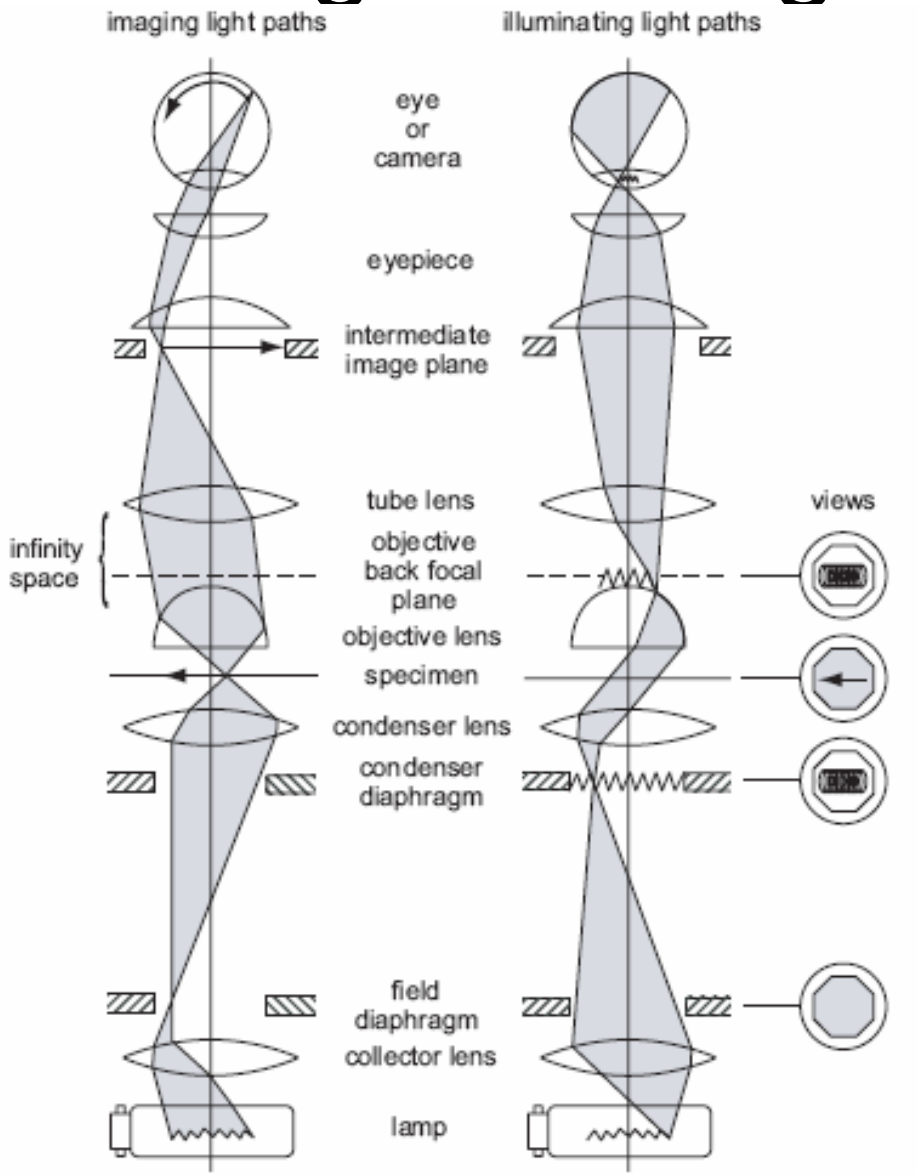
An image is always imperfect since a lens with a finite diameter captures limited frequency information

PSF is the Green's function for an optical system.
OTF is its Fourier Transform, characterizes a systems frequency response.

Microscopy (this lab)

- Brightfield
- Darkfield
- Phase contrast
- TIRF
- Optical tweezers
- Fluorescence (epi-illumination)

Brightfield light path



We can understand from this how to align the only movable element, the condenser for Kohler illumination.

$$R = \frac{1.22\lambda}{NA_{obj} + NA_{con}}$$

Darkfield

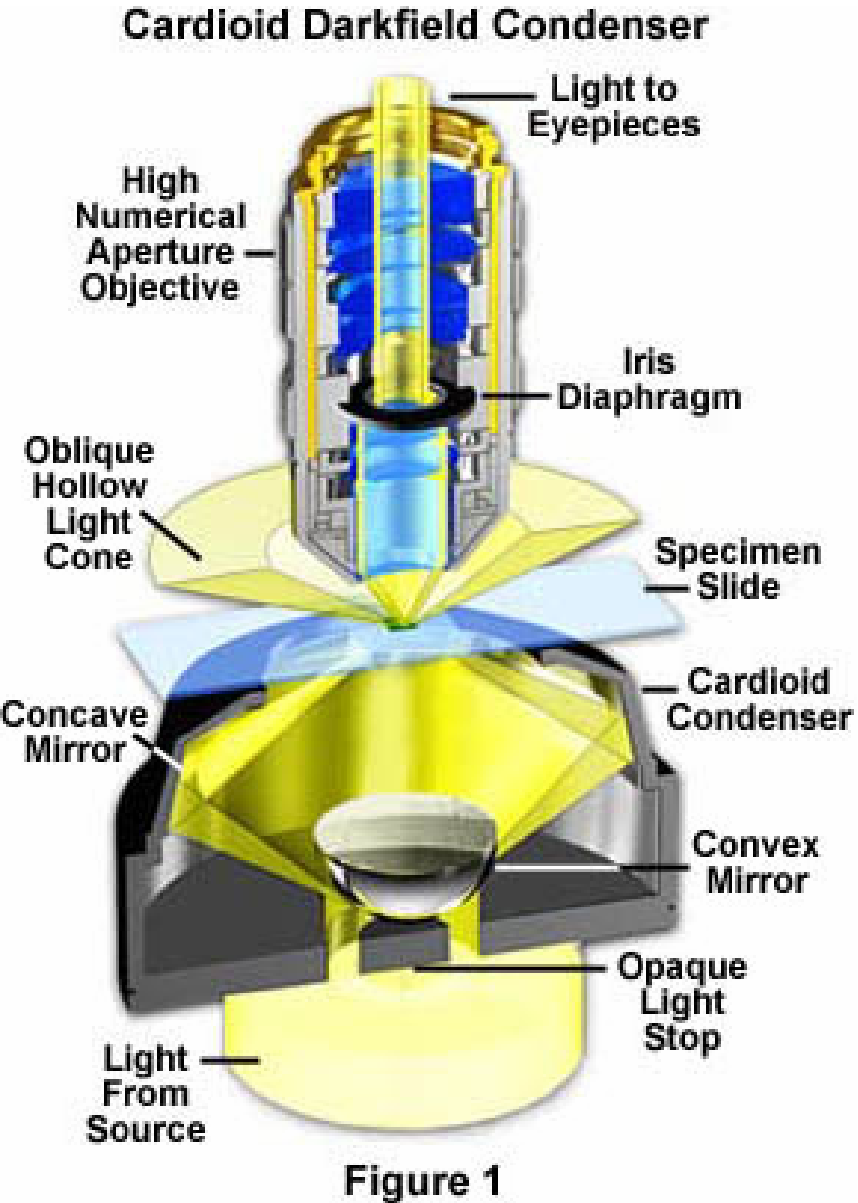


Figure 1

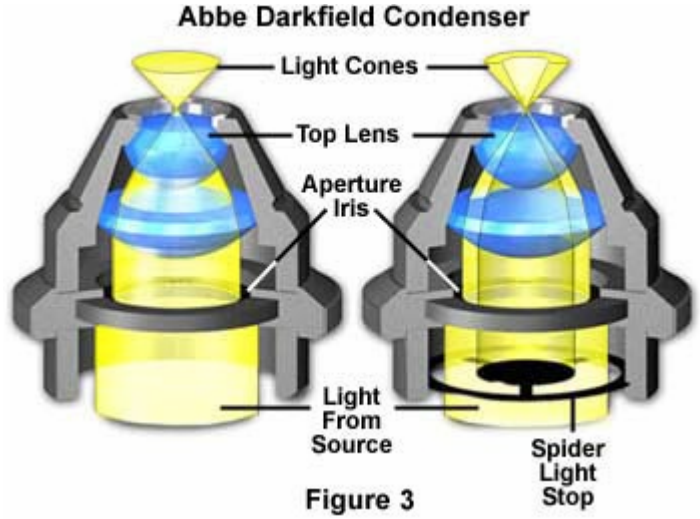


Figure 3

The condenser blocks out the 0th order light, only allowing higher orders to pass, enhancing contrast.

Phase contrast

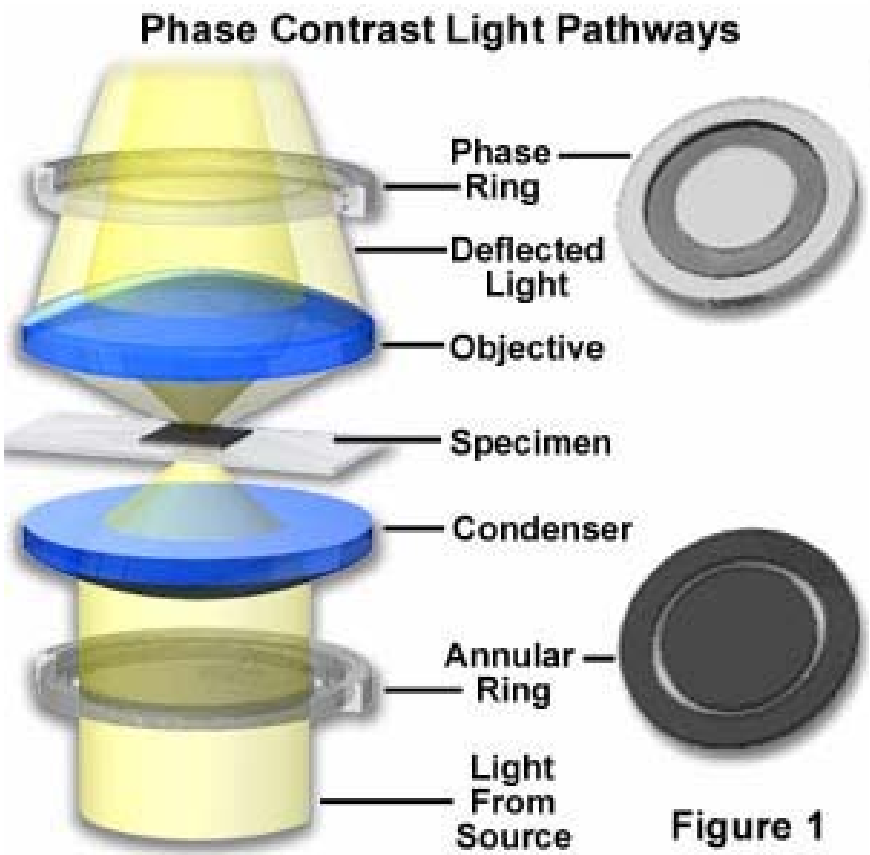
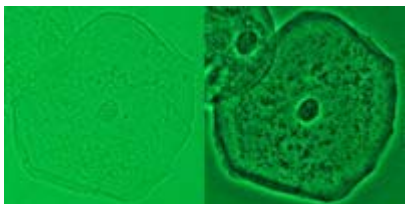


Figure 1



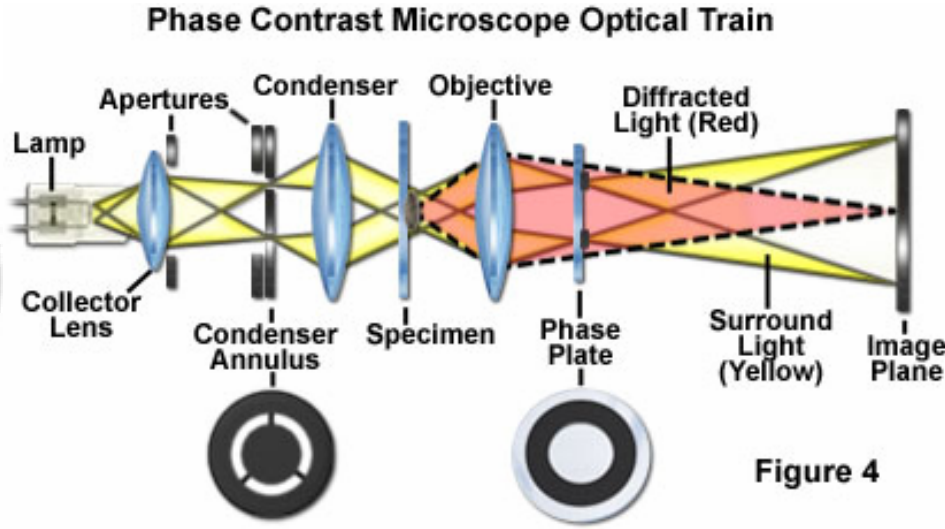
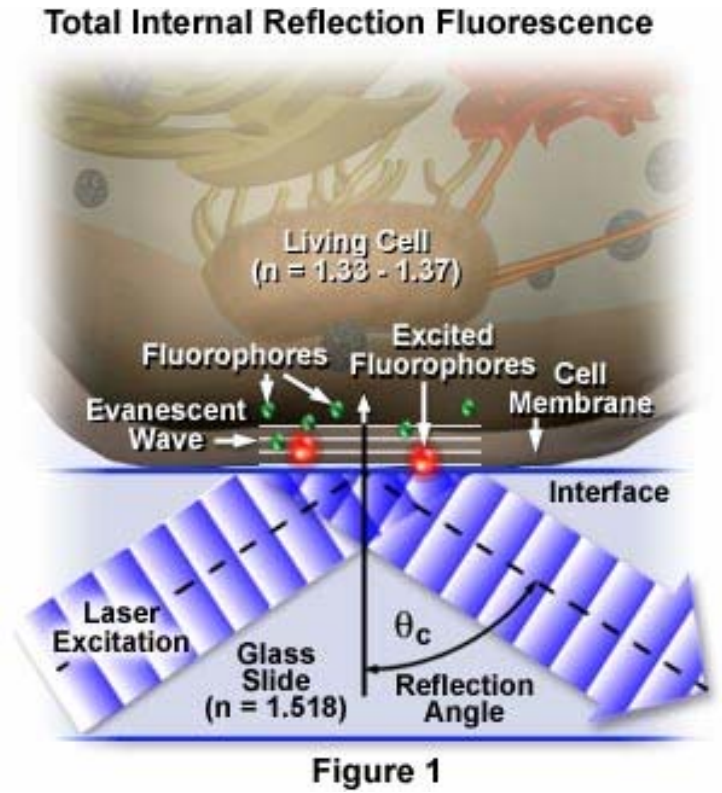


Figure 4

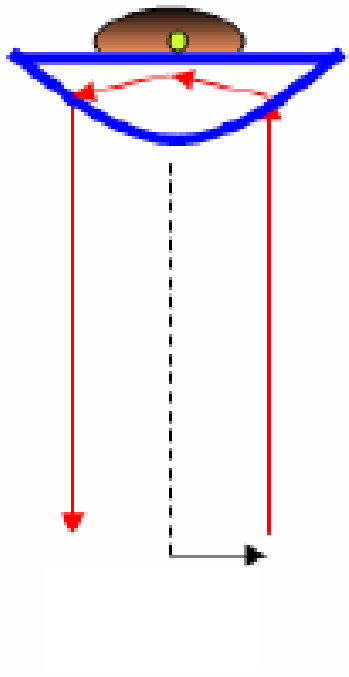
By “speeding up” the 0th order light by ¼ wave, it destructively interferes with the diffracted light (green).

This is an instance of Fourier plane filtering.

TIRF

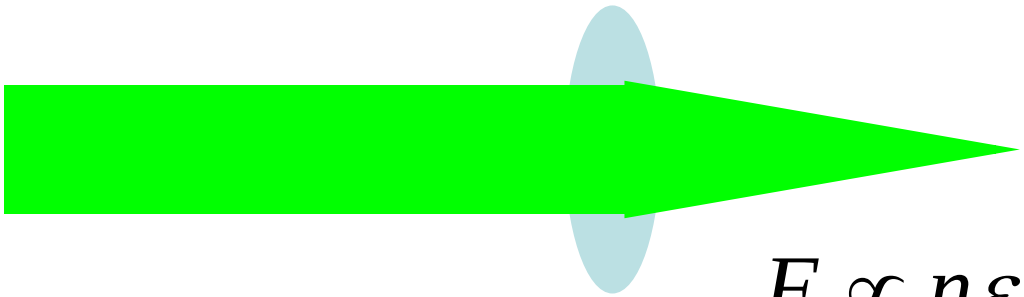


Snell's law



$$I(x) = I_0 \cdot e^{-\frac{X}{a}} \quad ; \quad \mathrm{a} = \frac{\lambda}{4\pi \sqrt{n_{\mathrm{glass}}^2 \sin^2 \alpha - n_{water}^2}}$$

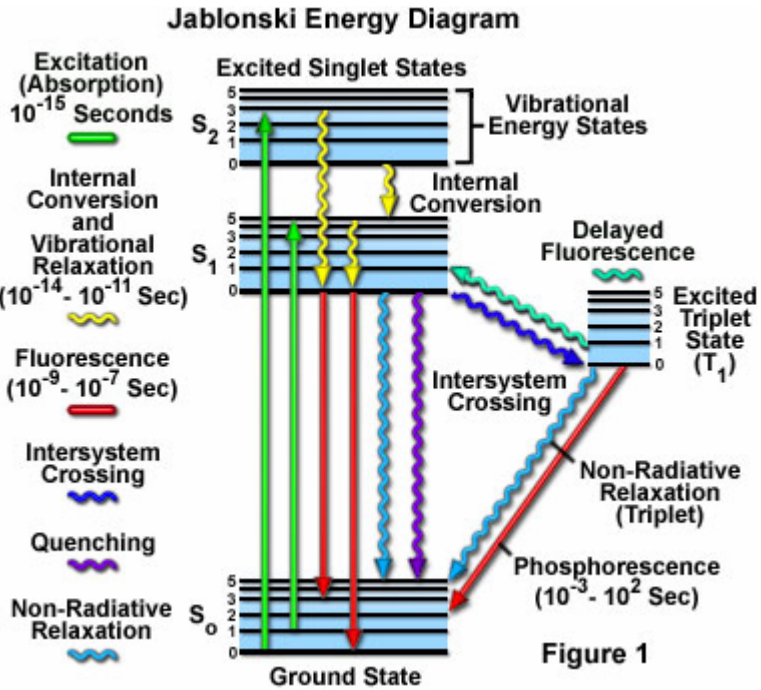
Optical tweezers

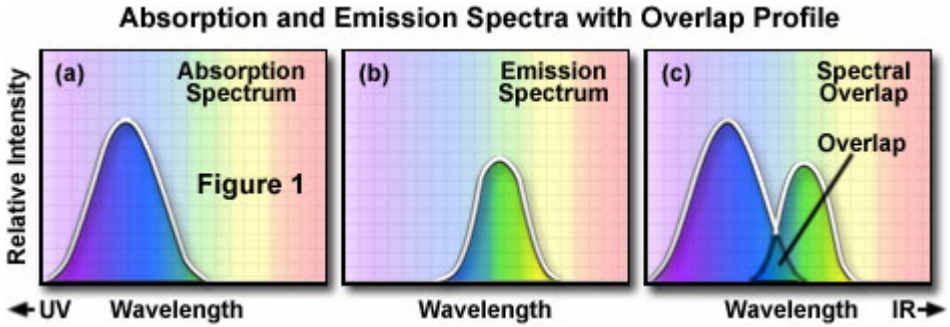


$$F \propto n\varepsilon_o E^2$$

Need high NA to achieve high enough intensity to trap stuff

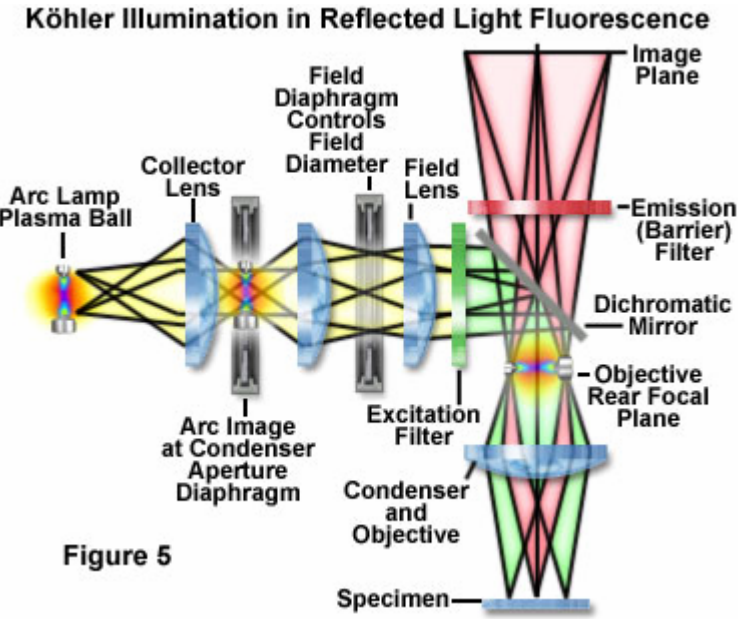
Fluorescence

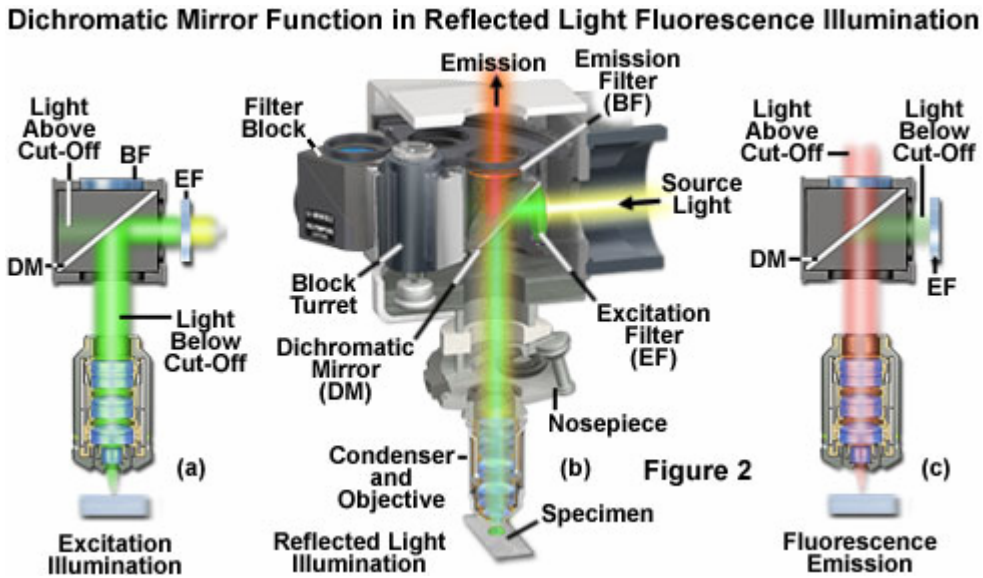




Fluorescence is an electronic state relaxation phenomenom
-Photobleaching
-FRAP
-FRET
-High resolution localization

Fluorescence





So… how do we align the arc lamp of a fluorescence microscope?