Optical methods in chemistry
or
Photon tools for chemical sciences

Session 7:

Course layout – contents overview and general structure

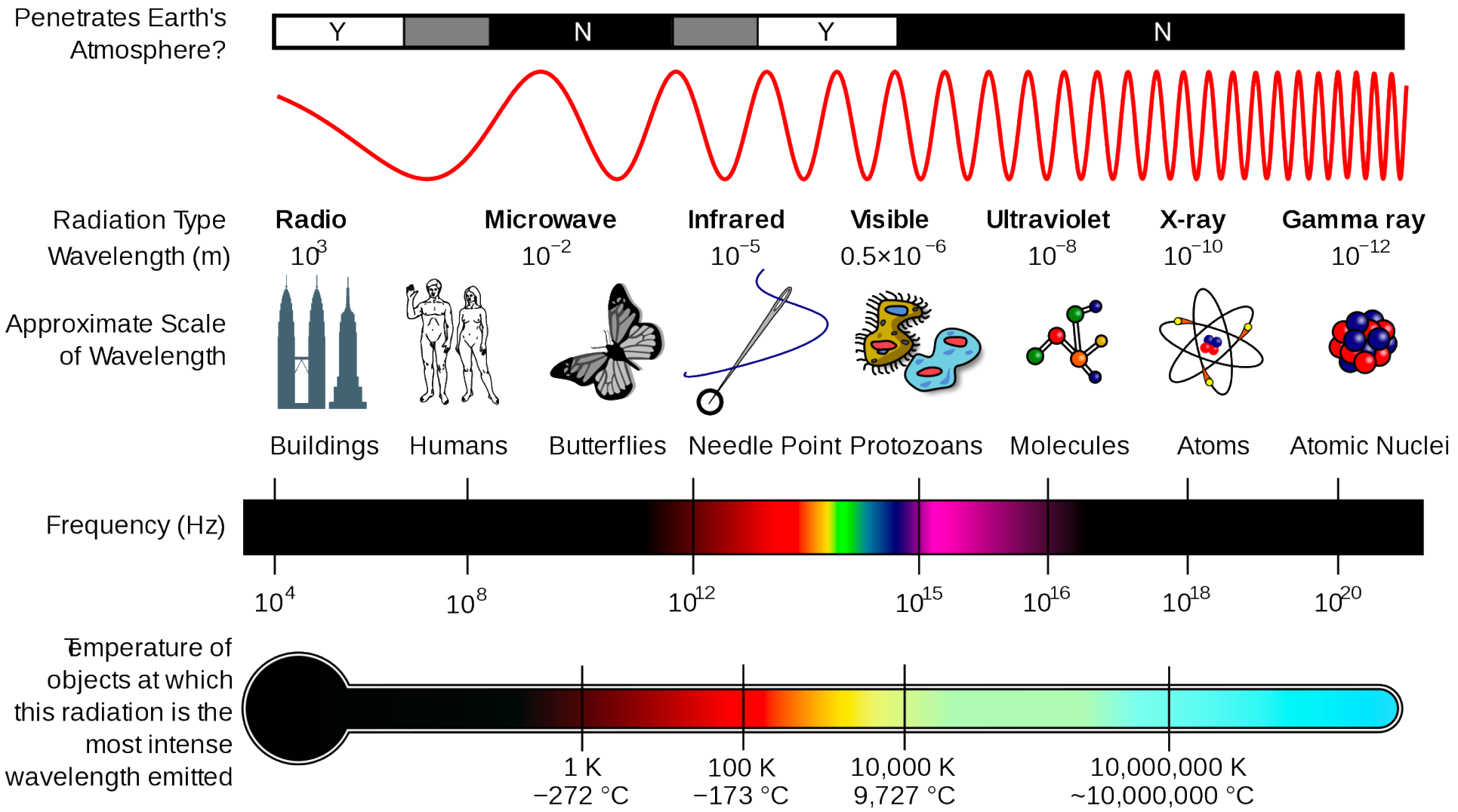
- Introduction and ray optics
- Wave optics
- Beams
- From cavities to lasers
- More lasers and optical tweezers
- From diffraction and Fourier optics
- **Microscopy**
- Spectroscopy
- Electromagnetic optics
- Absorption, dispersion, and non-linear optics
- Ultrafast lasers
- Introduction to x-rays
- X-ray diffraction and spectroscopy
- Summary

Going towards applications:

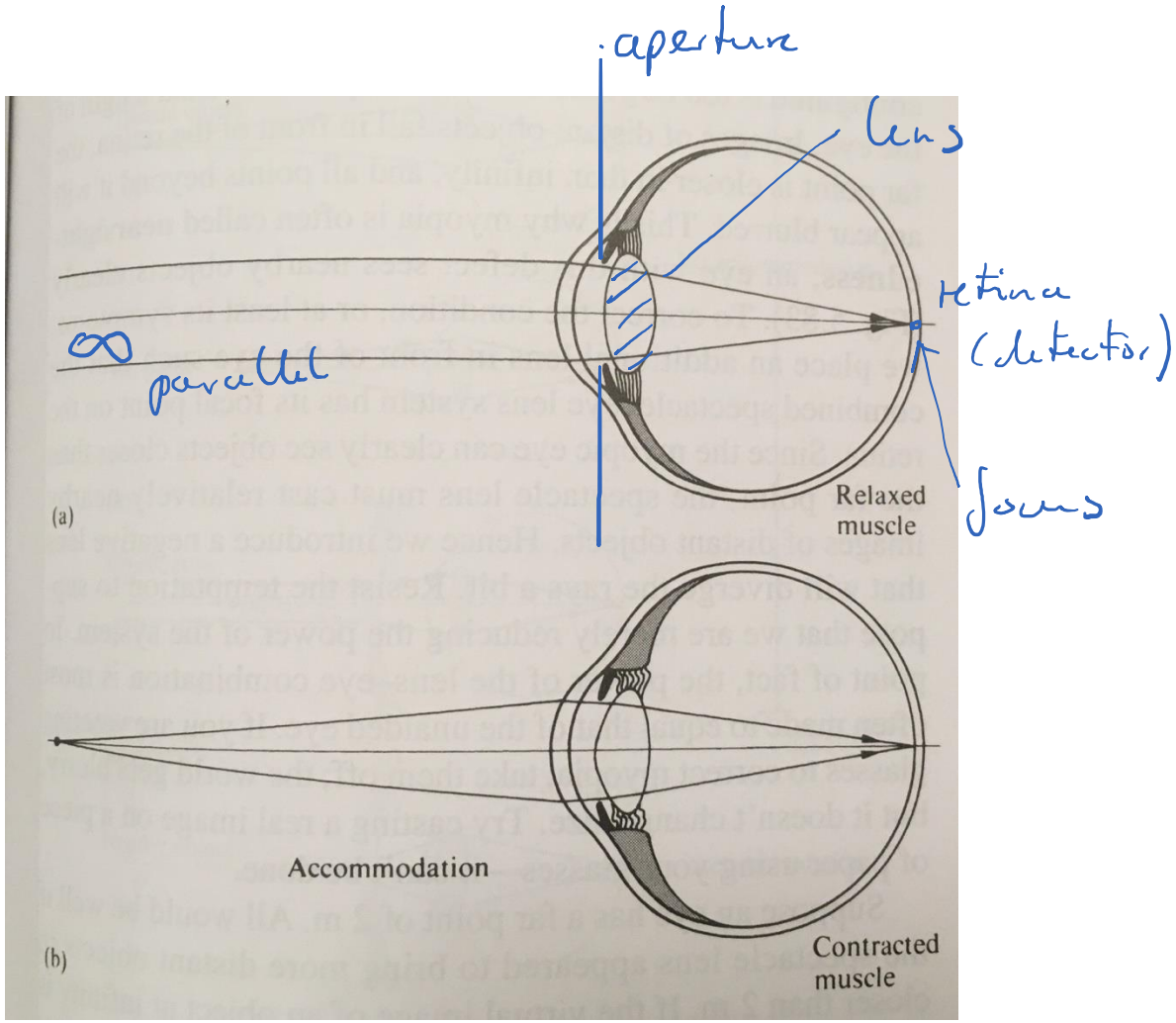
However, still focus on some concepts

Microscopy – Manipulation - Spectroscopy

Remember: Energy – wavelength – size relationship



Reminder: Our eye as an optical system



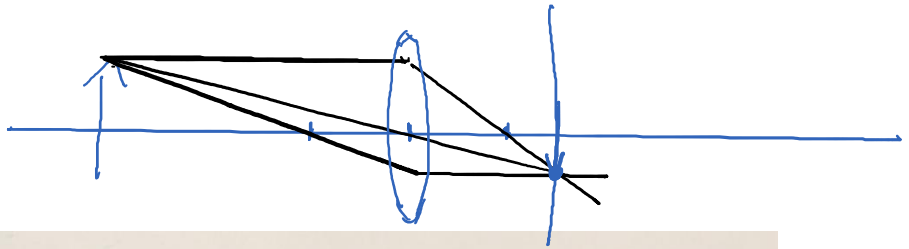
This makes laser radiation so dangerous to eye: parallel beams give perfect focus

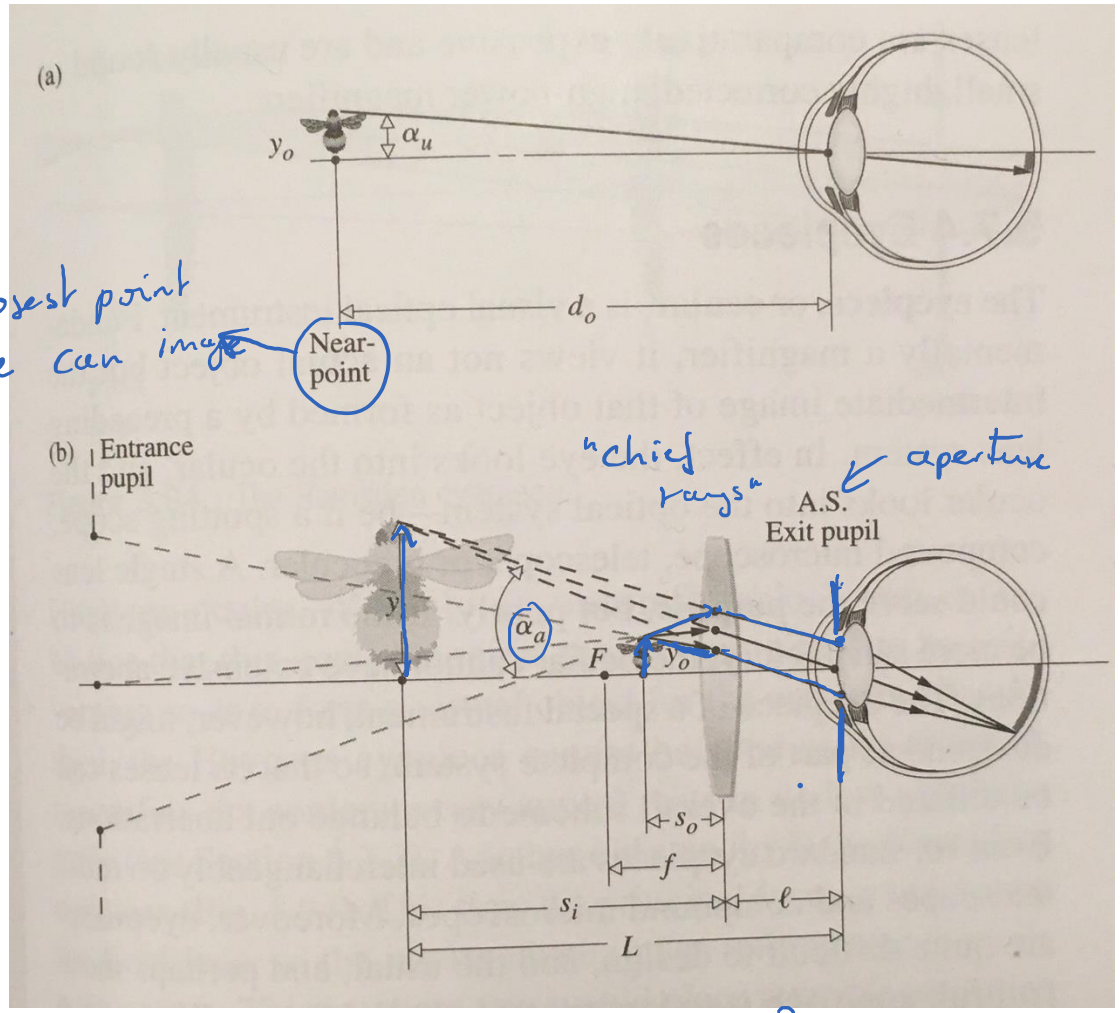
When designing optical systems (microscopes) need to consider eye as optical element.

Nowadays eye is typically replaced by camera which is also an imaging system.

Magnifying glas: a simple optical element

eye

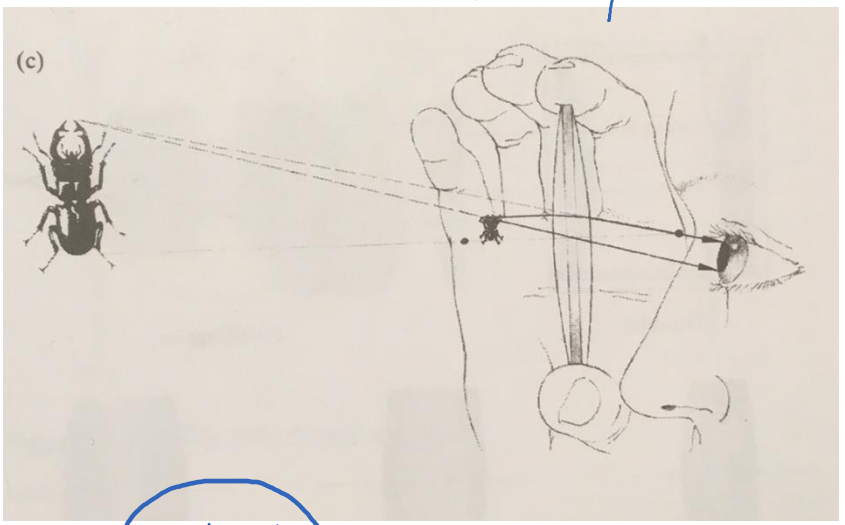


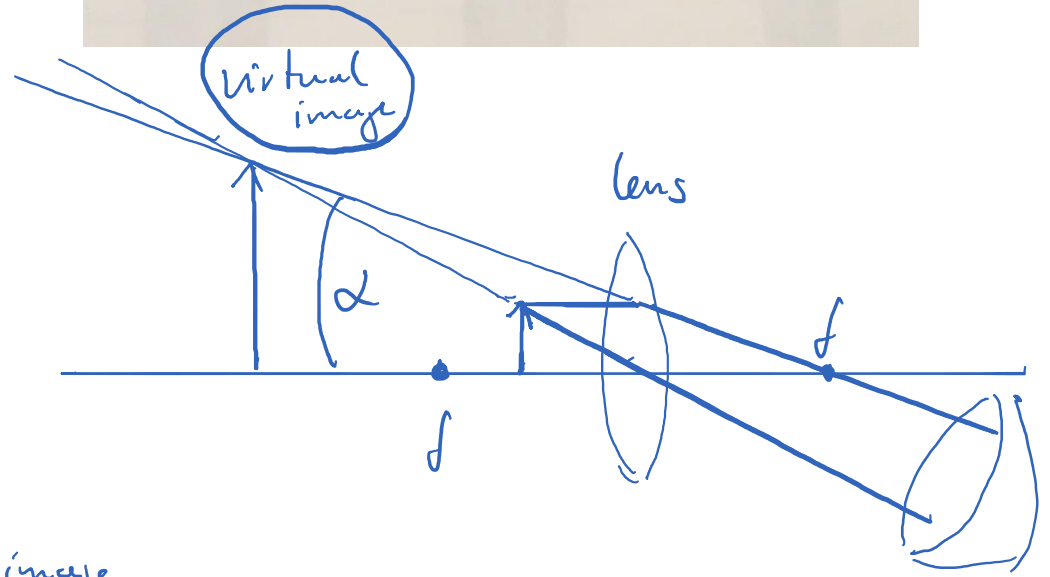


closest point eye can image

"chief rays"

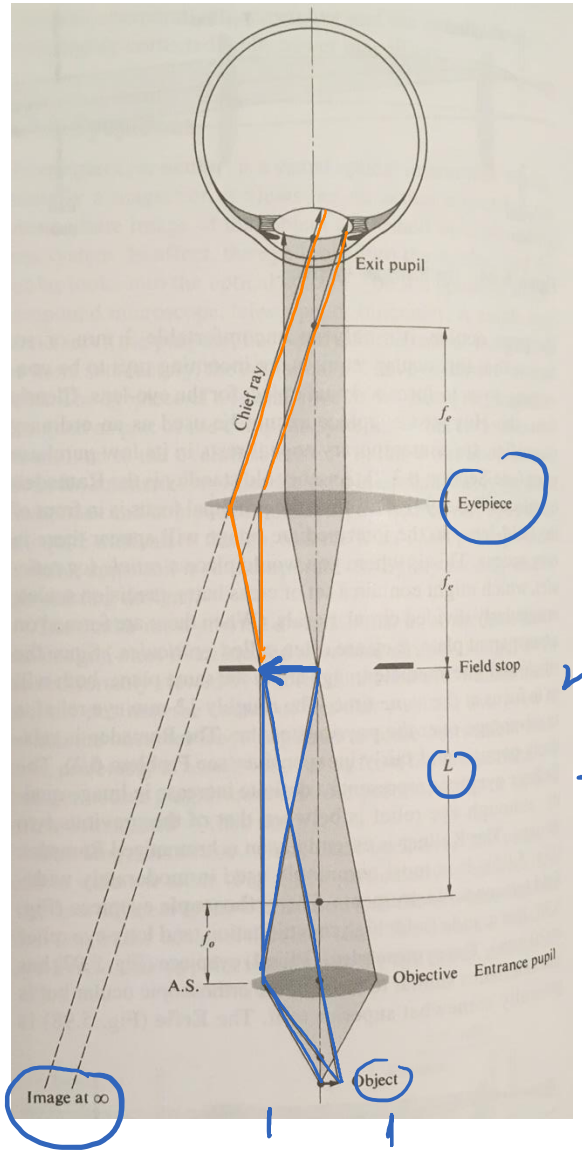
aperture





angular magnification $M = \frac{\alpha_a}{\alpha_u}$ } size of retinal image of instrument over unaided eye

The microscope – a classical view



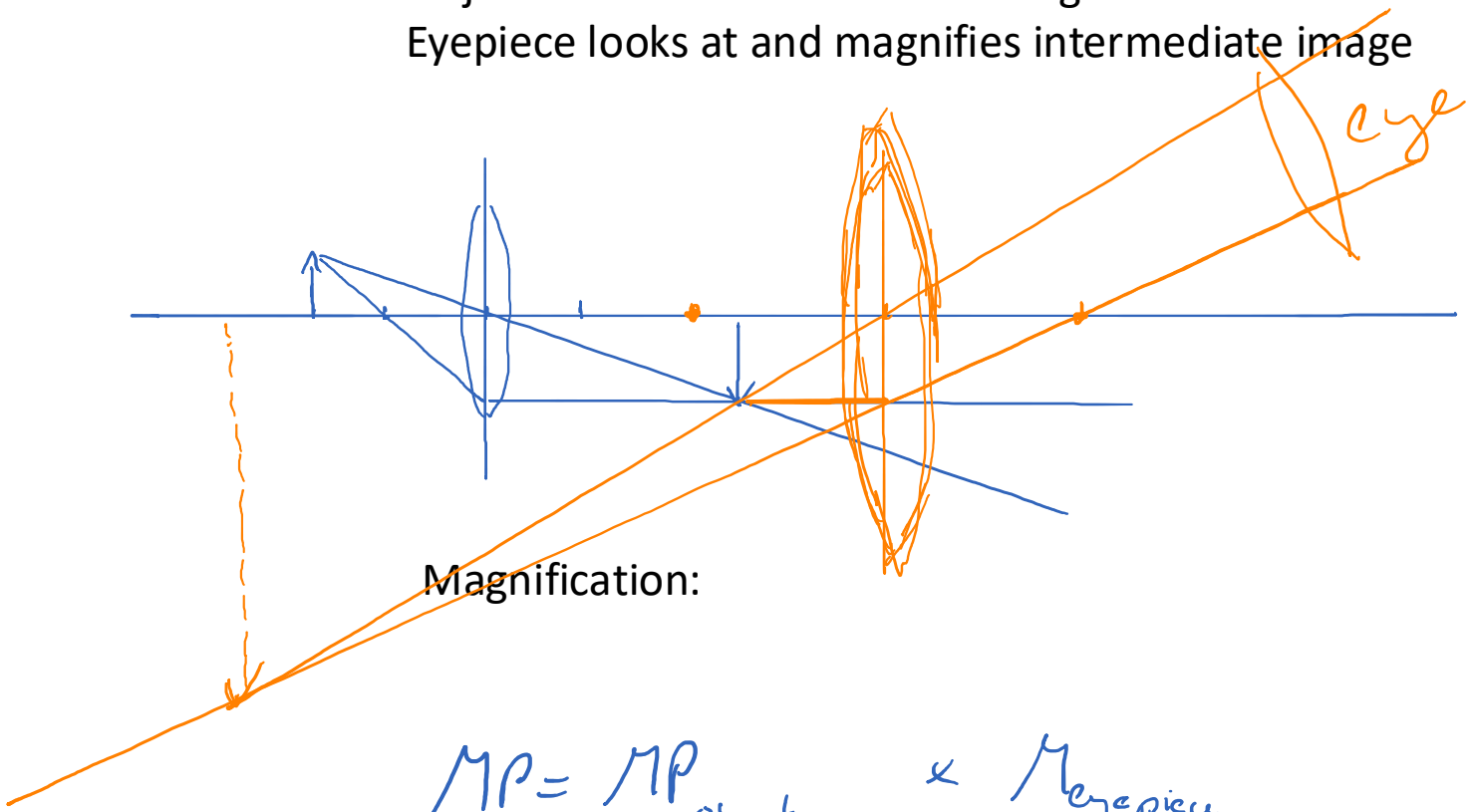
magnifying glass

real image

tube length convention ~160 mm

size of microscope

Principle:
Objective forms intermediate image
Eyepiece looks at and magnifies intermediate image



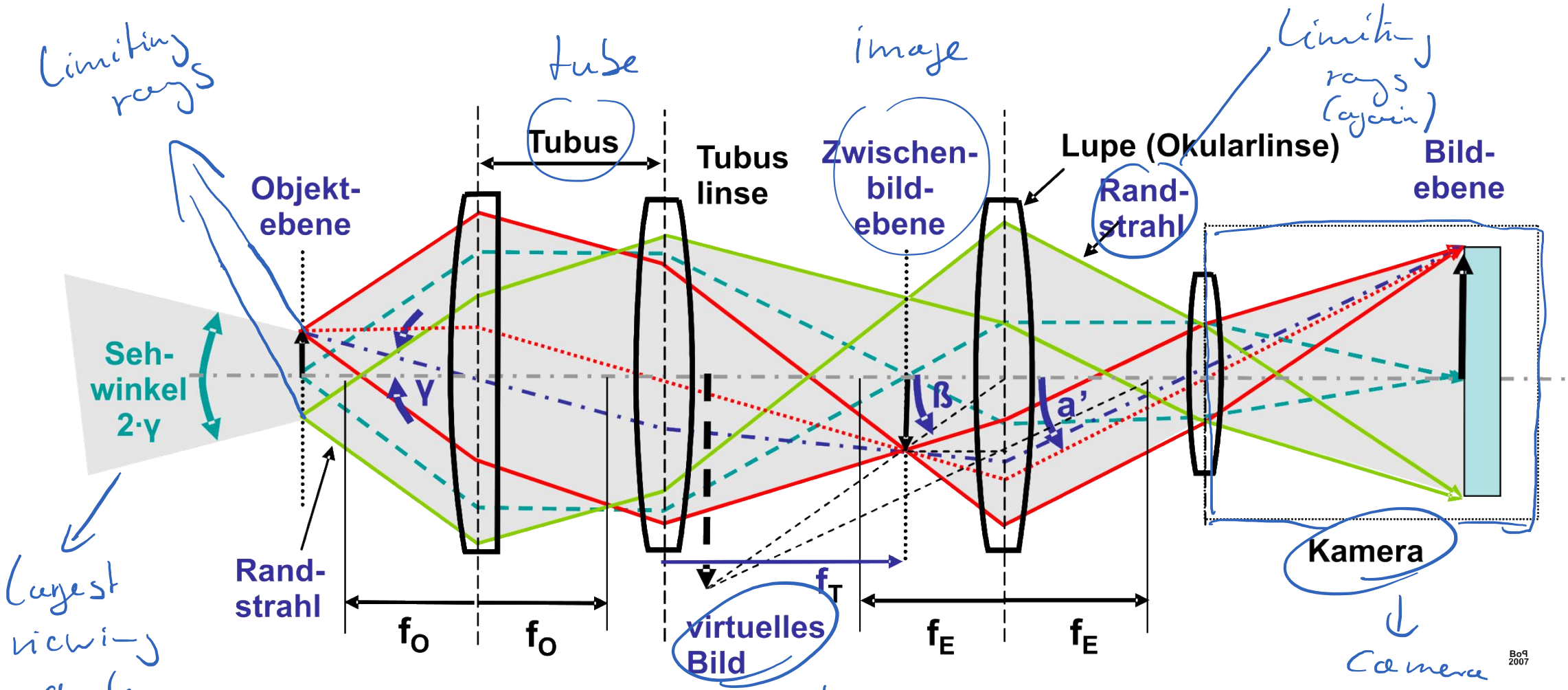
Magnification:

$MP = MP_{objective} \times M_{eyepiece}$

$\sim 50 \qquad \sim 10$

500 !

More extensive and more realistic schematic:



↳ virtual image

Note 2nd Linse in tube for parallel beam → optical components → "infinity corrected" microscope

Microscope variations:

There are many variations and combinations with other techniques for microscopes in terms of

- Illumination ← So Hum / top
- Magnification ← Lens systems
- Detectors ← eye, camera, ...
- Staining for improving contrast
- Fluorescence detection
- Etc

Lets look at some conceptual variations

Microscope variations: Confocal microscope

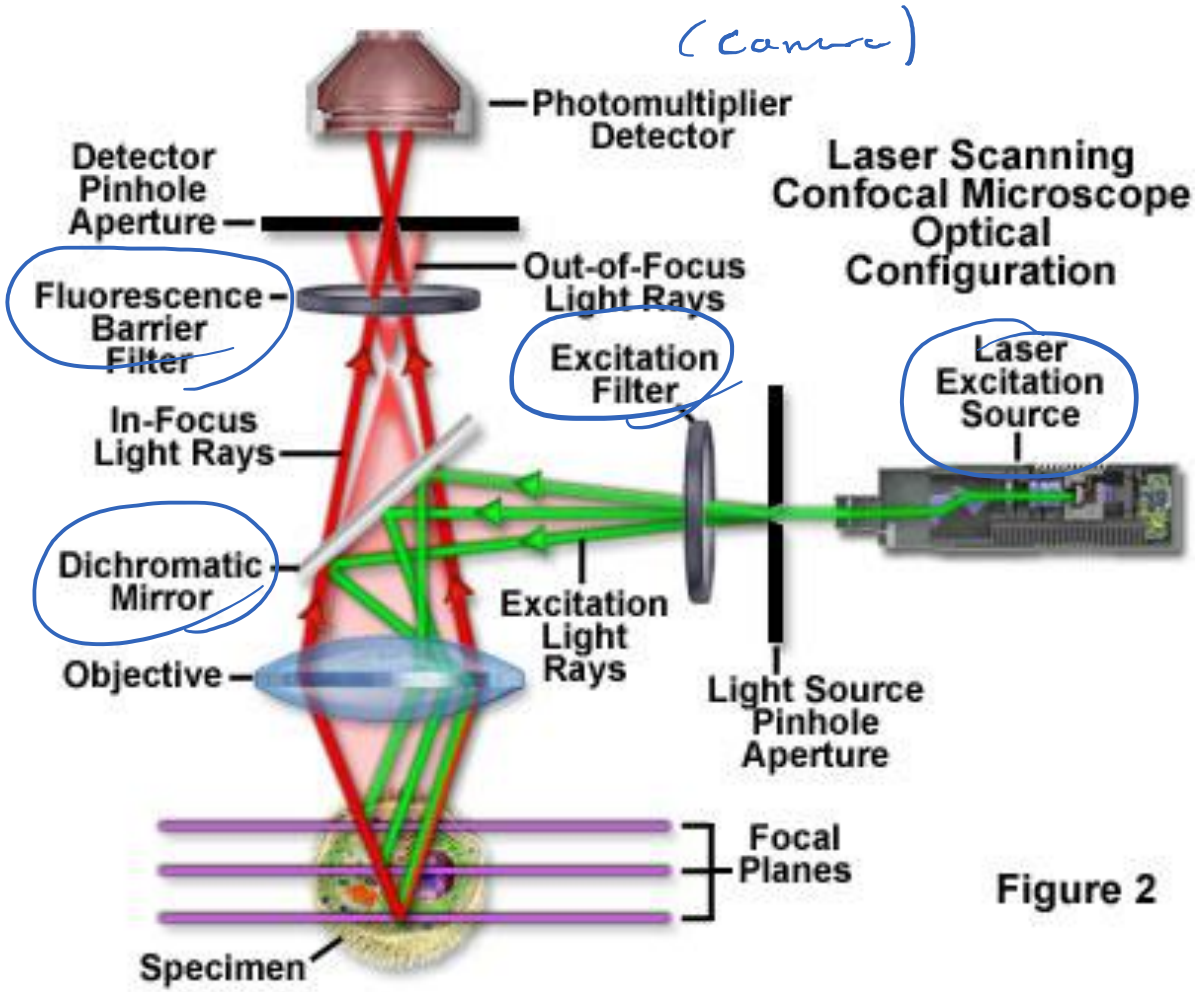
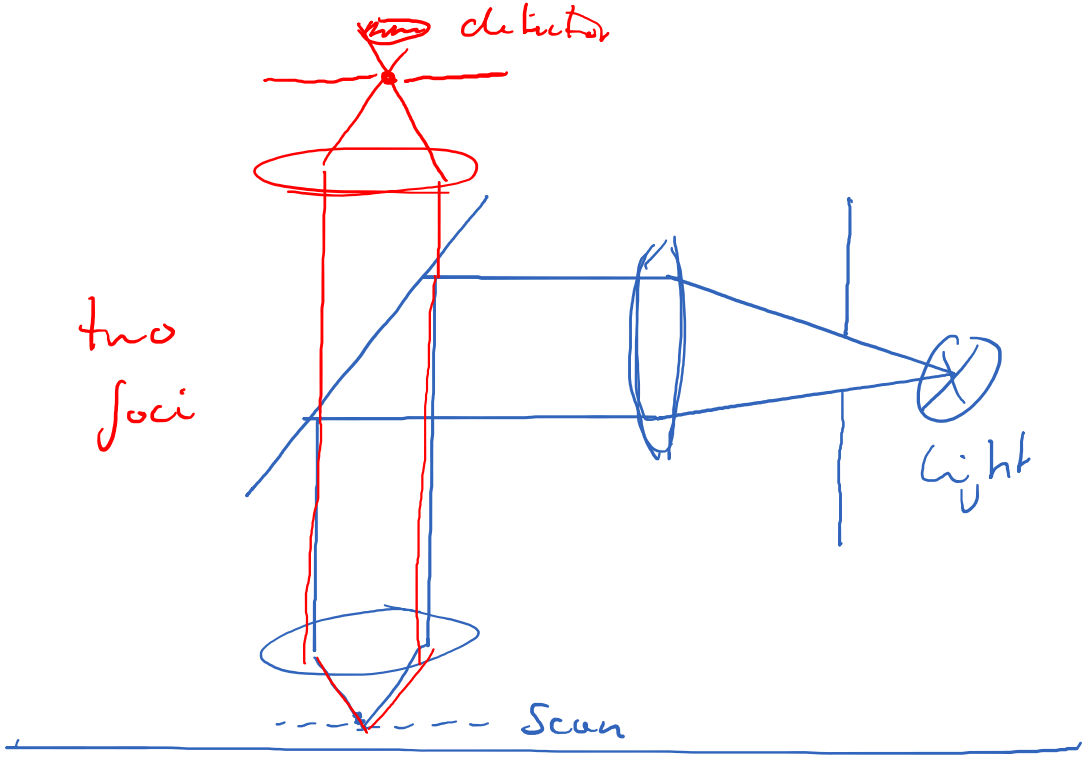


Figure 2

Source: Olympus



→ not full image but scan

→ illuminate small part

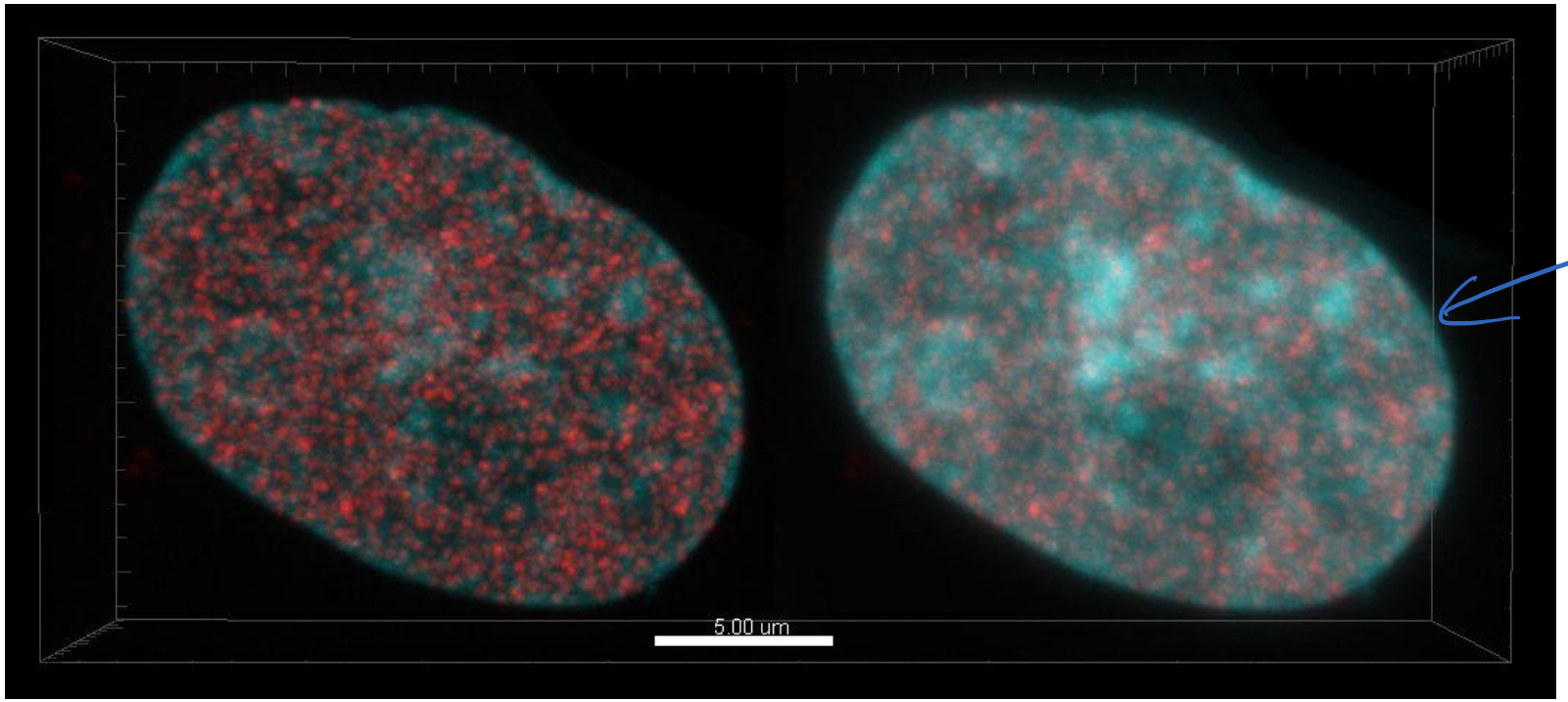
→ aperture & light point in focus
↳ confocal

→ allow only light from focus to detector

→ Scan sample
→ reconstruct in computer
} 3D info!

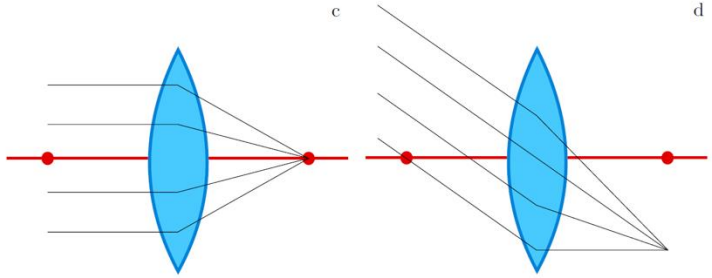
Confocal microscopy: Example

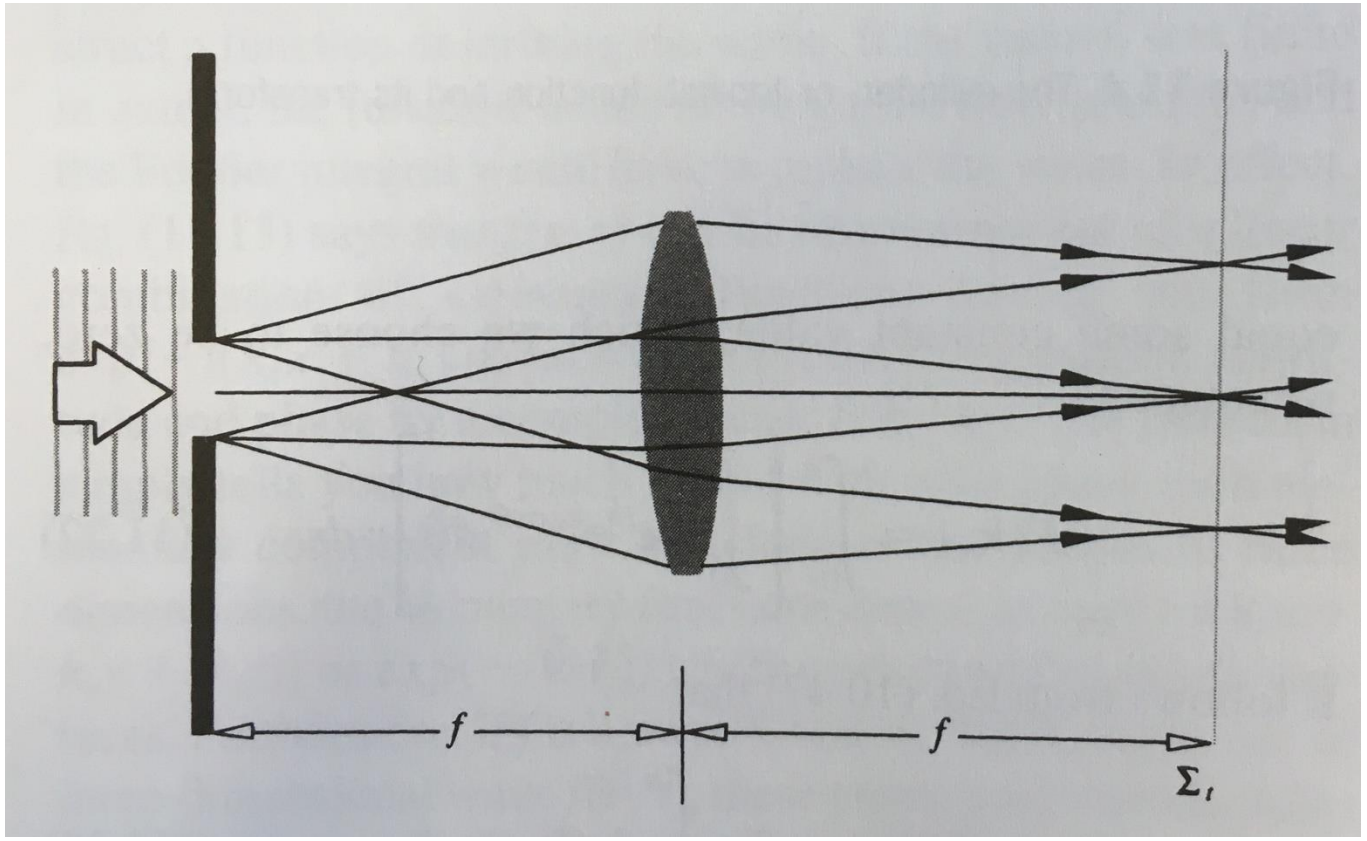
- Optical cuts through cell nucleus with two different pinhole / aperture sizes
- Computer can generate 3D models. Fluorescence excitation can be used to highlight specific units
- https://upload.wikimedia.org/wikipedia/commons/9/92/Combined-confocal-unconfocal.gif



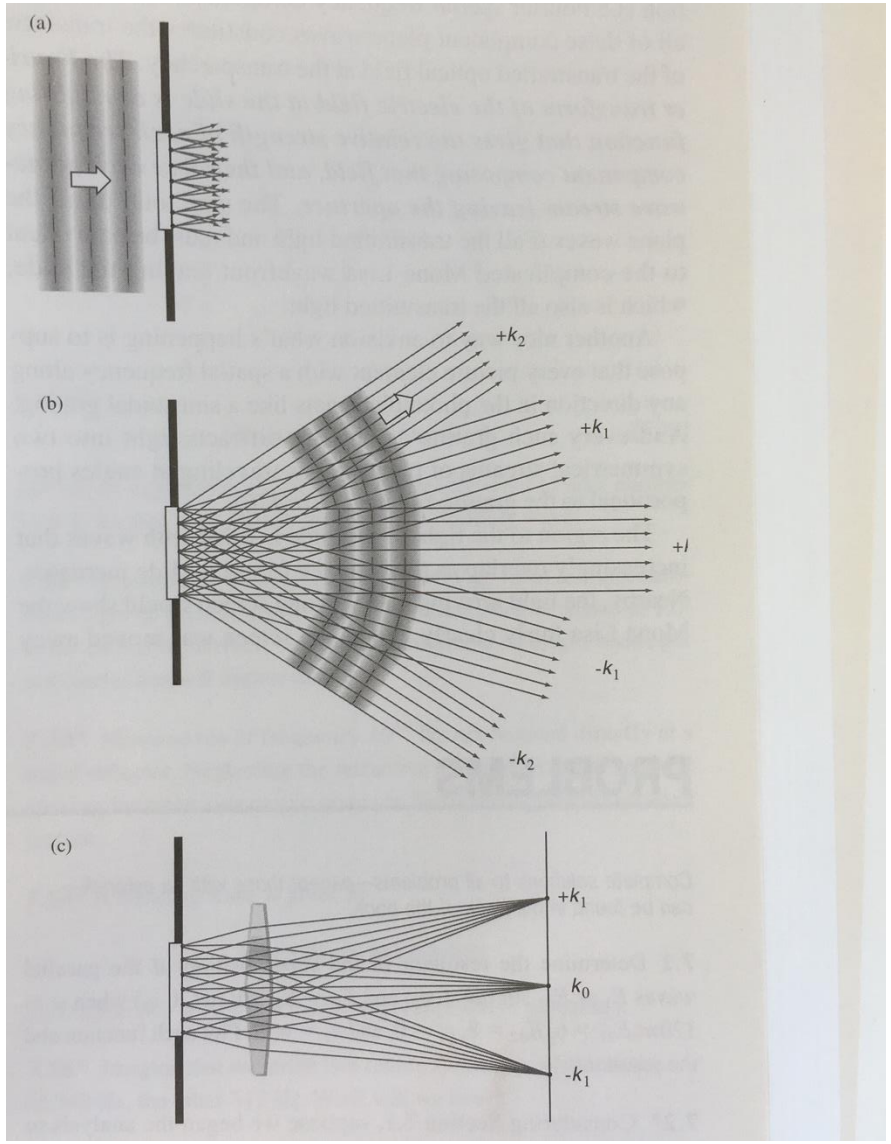
Lens as Fourier transformer

Remember!





Gedankenexperiment for imaging: (repeat)



imaging as diffraction

lens "pulls" diffraction pattern in Fourier plane

Abbe image formation

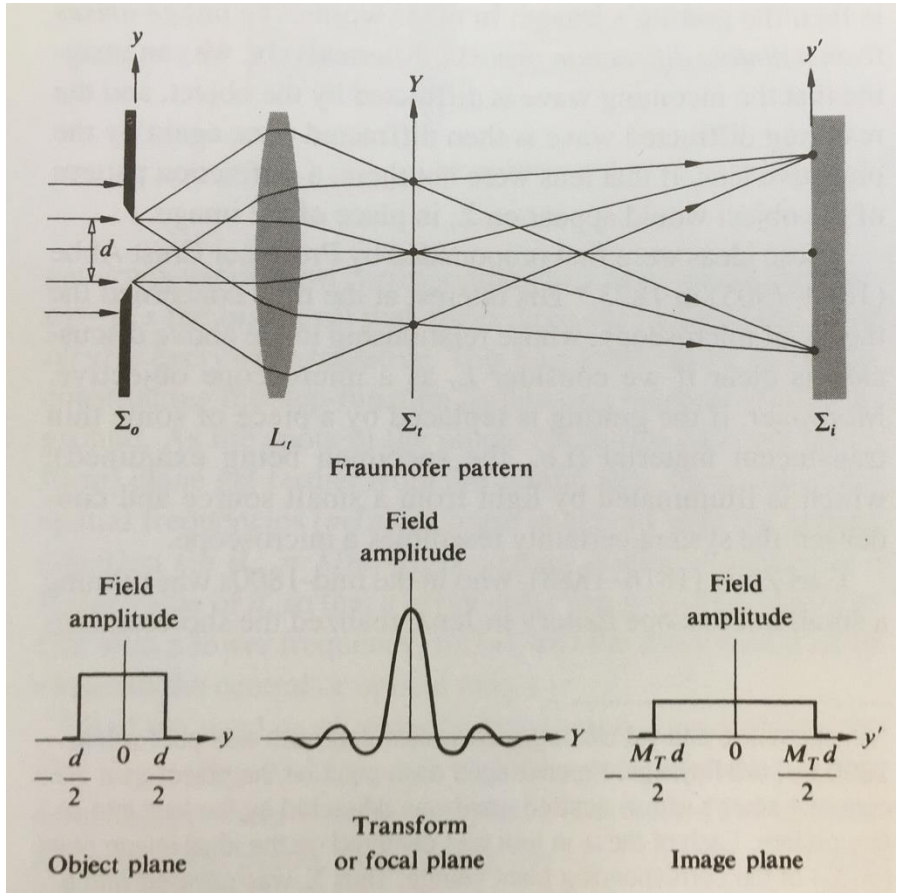


image formation as diffraction process

↓

Light waves are diffracted by the object

↓

diffraction pattern (FT of object) is formed in focal plane

↓

importance to capture higher orders by objective lense

Phase contrast microscopy

→ use phase ↔ interference

→ use when absorption is weak

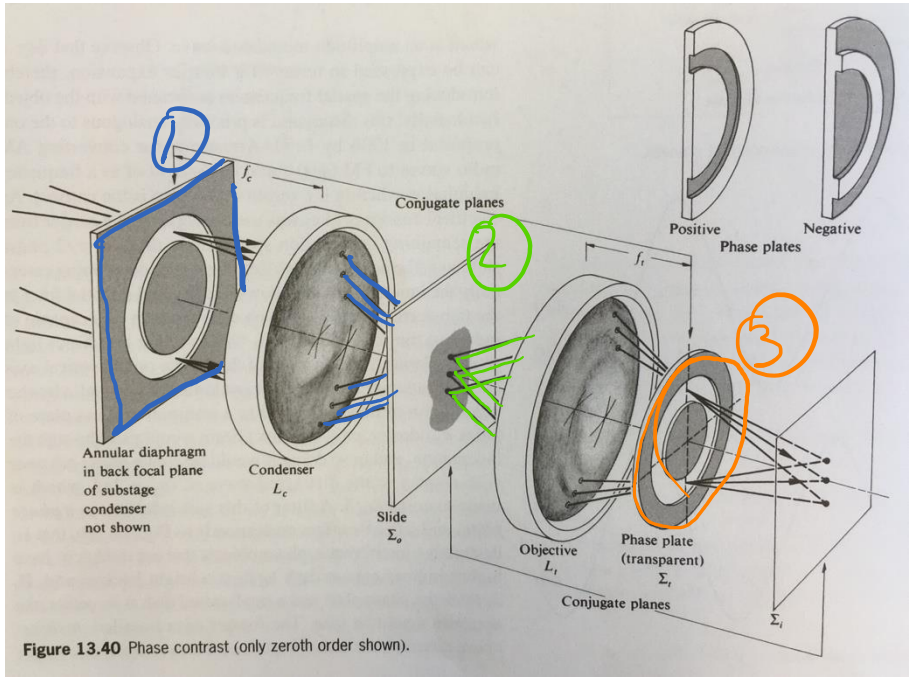
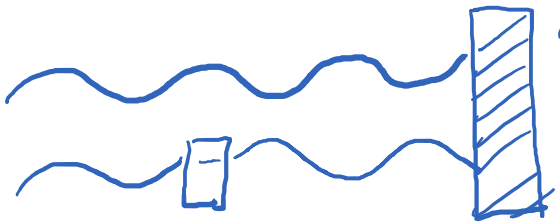


Figure 13.40 Phase contrast (only zeroth order shown).



detection ↓ interference

The Nobel Prize in Physics 1953

Photo from the Nobel Foundation archive.

Frits Zernike

Prize share: 1/1

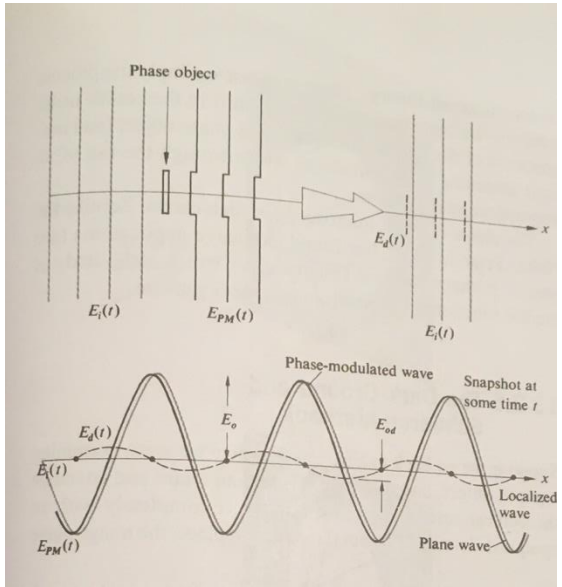
→ detect interference

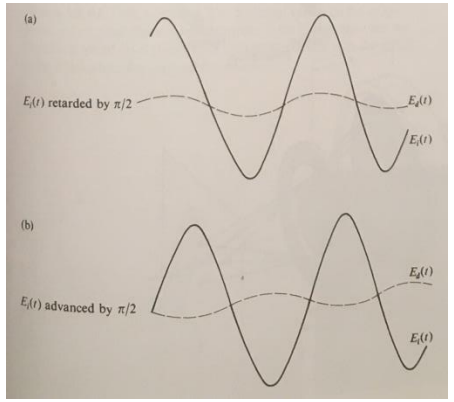
→ change detection from amplitude to interference

① Spatially defining aperture

② Light is scattered in sample (weak)

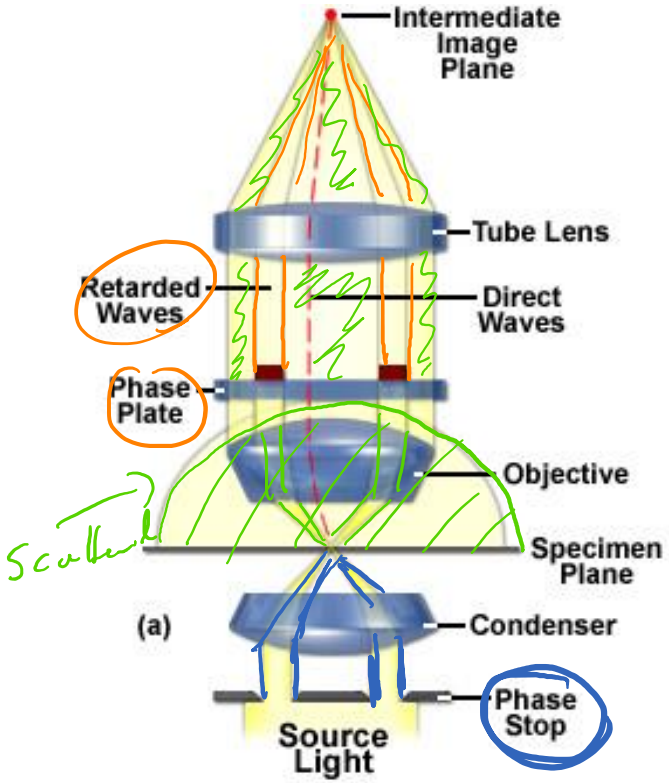
③ phase plate attenuates & phase shift direct rays

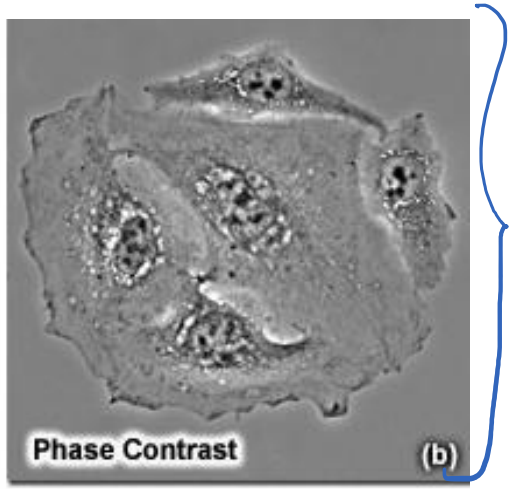


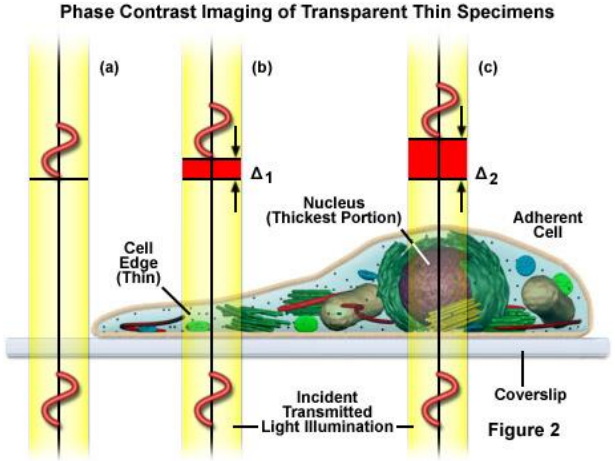


⇒ art is to fine-tune phase / amplitude for best contrast

Phase contrast microscopy



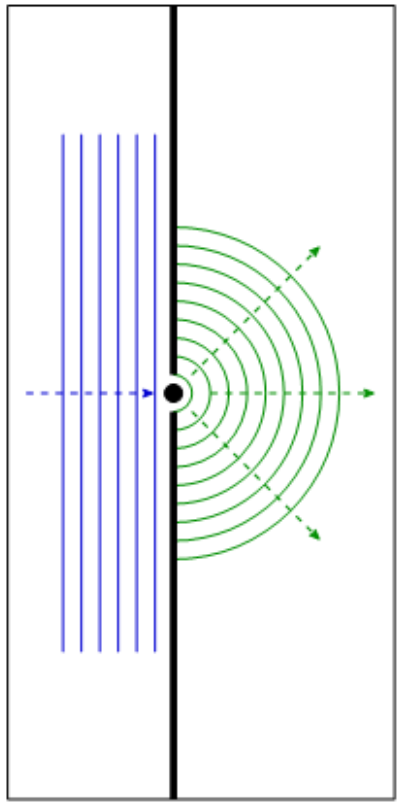




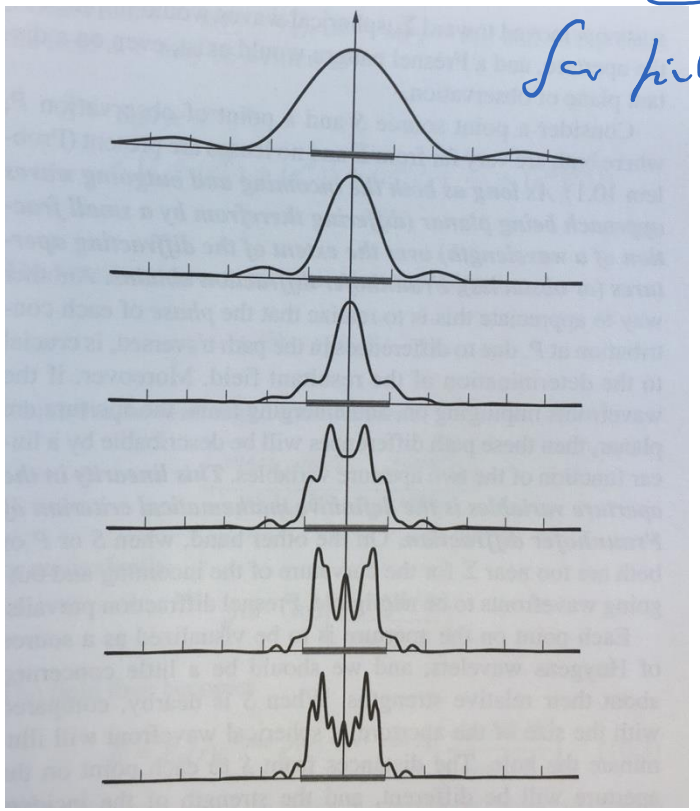
Source: Zeiss Campus

Resolution limit of a classical optical apparatus: Remember Huygens Fresnel principle, diffraction at a slit, Airy rings

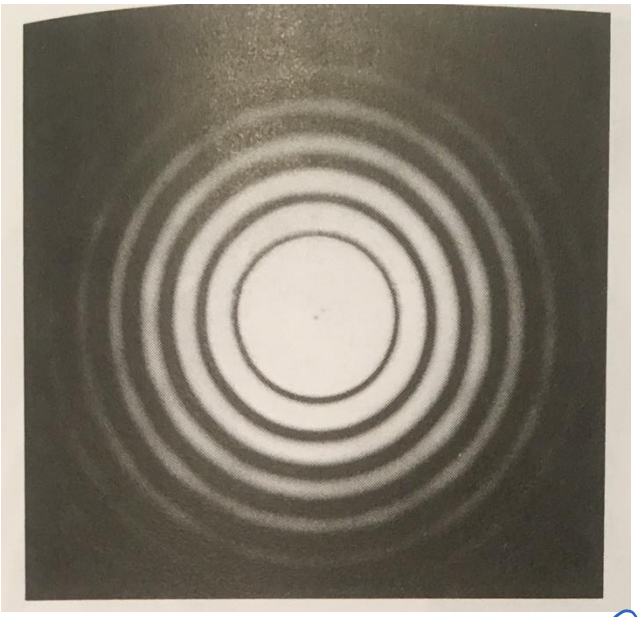
Diffraction at slit / aperture



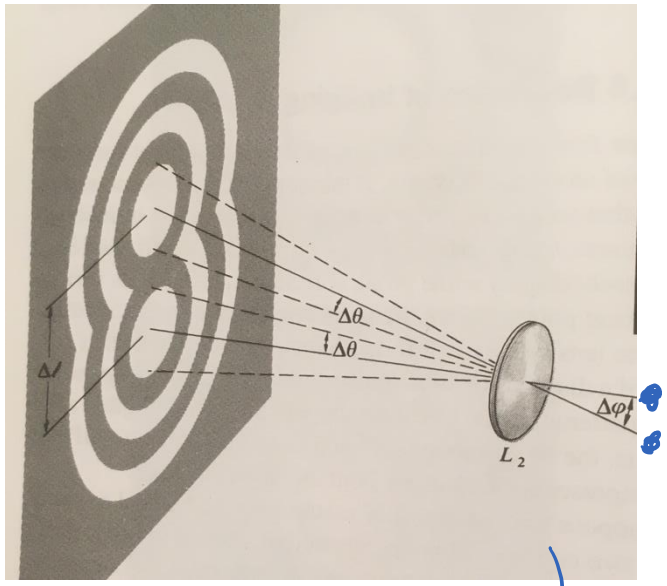
Fresnel and Fraunhofer regime



Airy rings of circular aperture



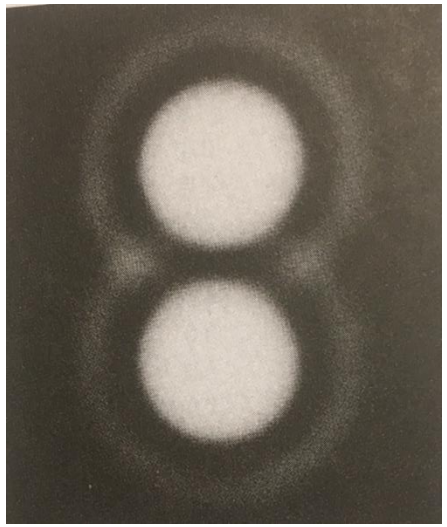
Resolution limit of a classical optical apparatus



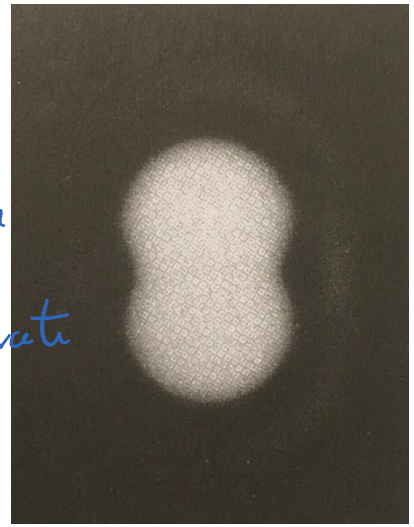
→ diffraction

two point sources

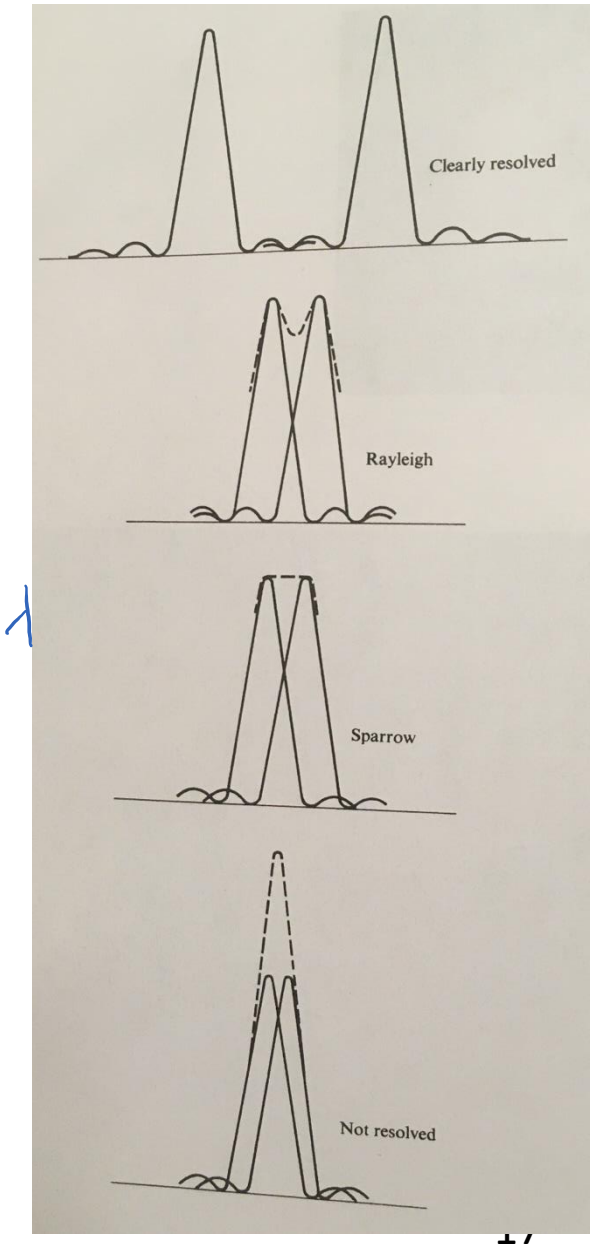
- limit of resolution for two point sources
- → size of Airy disks
- $\sim 1.22 \cdot \frac{f}{D} \cdot \lambda$
- → angle $\Delta\varphi = 1.22 \cdot \frac{f}{D} \lambda$



can separate

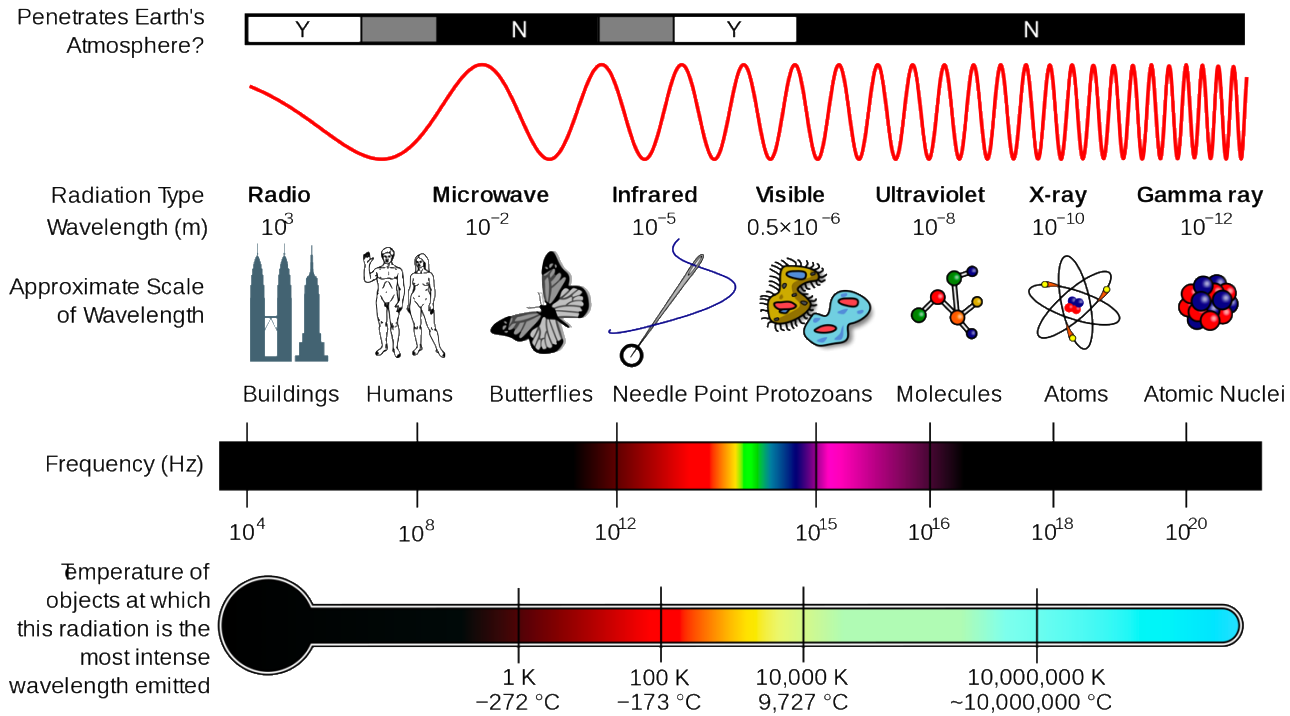


can not separate



How to beat resolution limit?

→ limiting factor is λ



Shorter wavelength:

X-ray microscopy. No optics available. Later in course.

Electron: wave nature or particles

Smart ideas?

De Broglie: all matter has particle and wave nature!

- 1929 Nobel prize
- Example: Electrons

$$\lambda_{particle} = \frac{h}{p} = \frac{h}{mv}$$

↳ electrons with velocity → λ

↳ velocity through potential difference

↳ high voltage

↳ example: 100 V (need to do relativistically)

$U = 100\,V \Rightarrow \lambda_{electron}$ 3.7 pm

→ high resolution imaging

Electron microscopy

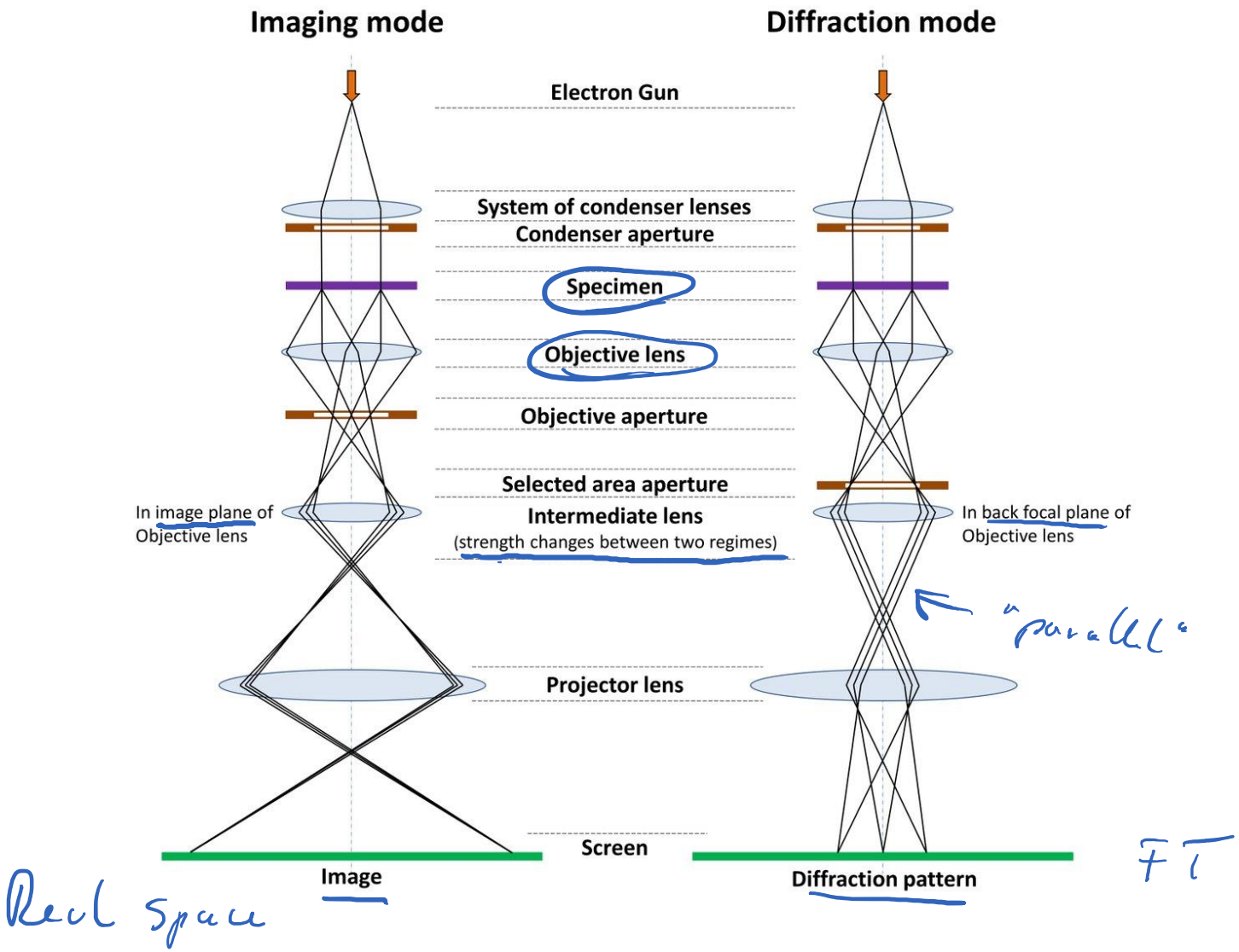
Electrostatic lenses akin optical lenses

Use short wavelength of (fast) electrons

Strong interaction with matter: Thins samples

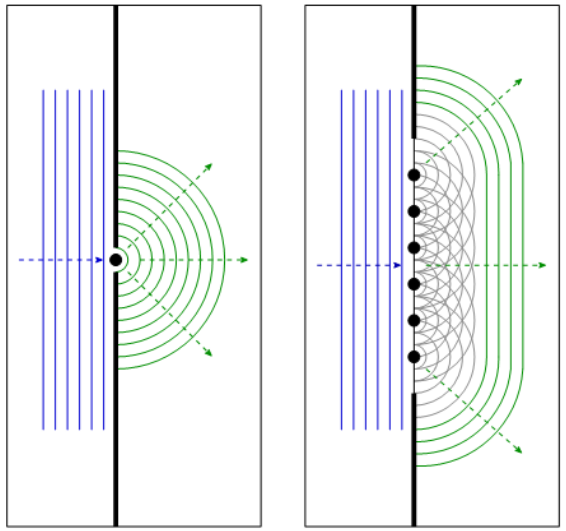
Need to use diffraction, understand Fourier planes, etc

This is an entirely different course but you need to know the close relationship with all optical concepts we have discussed

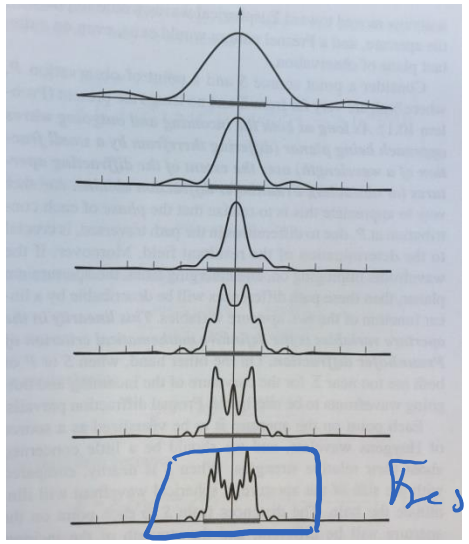


Overcoming the resolution limit: Near field approaches OR optics on the nanoscale

Small vs. real slit



Fresnel and Fraunhofer regime



↳ work close to sample

↳ scan aperture

Near field approach

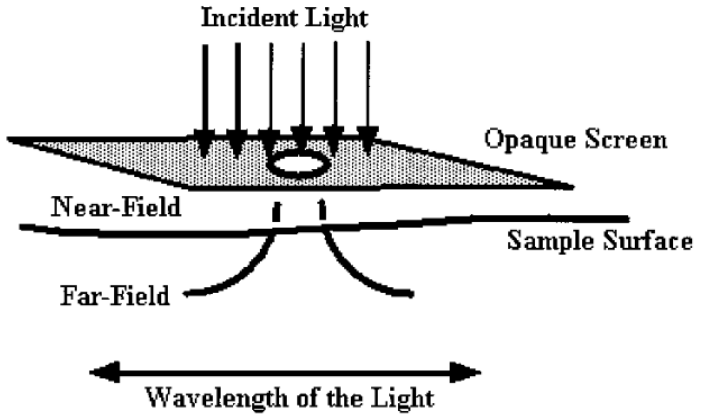


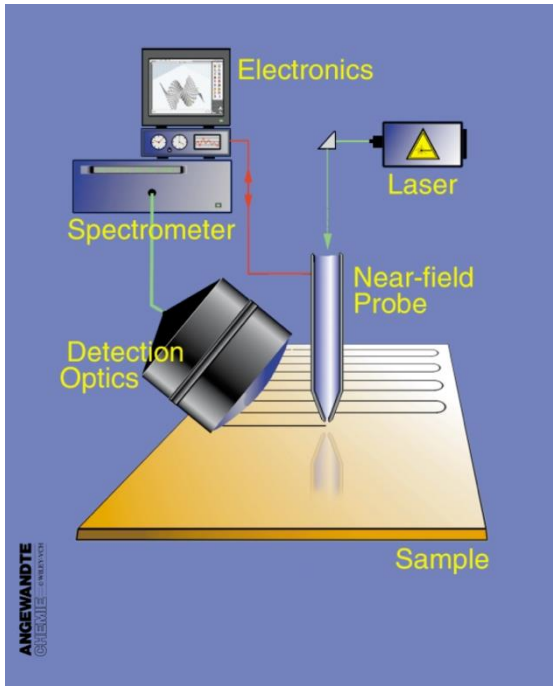
Figure 1. Schematic representation of Synge's idea for achieving subdiffraction limit spatial resolution. Incident radiation is passed through a small, subwavelength hole in an opaque screen. By the positioning of the screen close to the sample, the emerging radiation is forced to interact with the sample before diffracting out.

From: Dunn, Chem. Reviews 1999

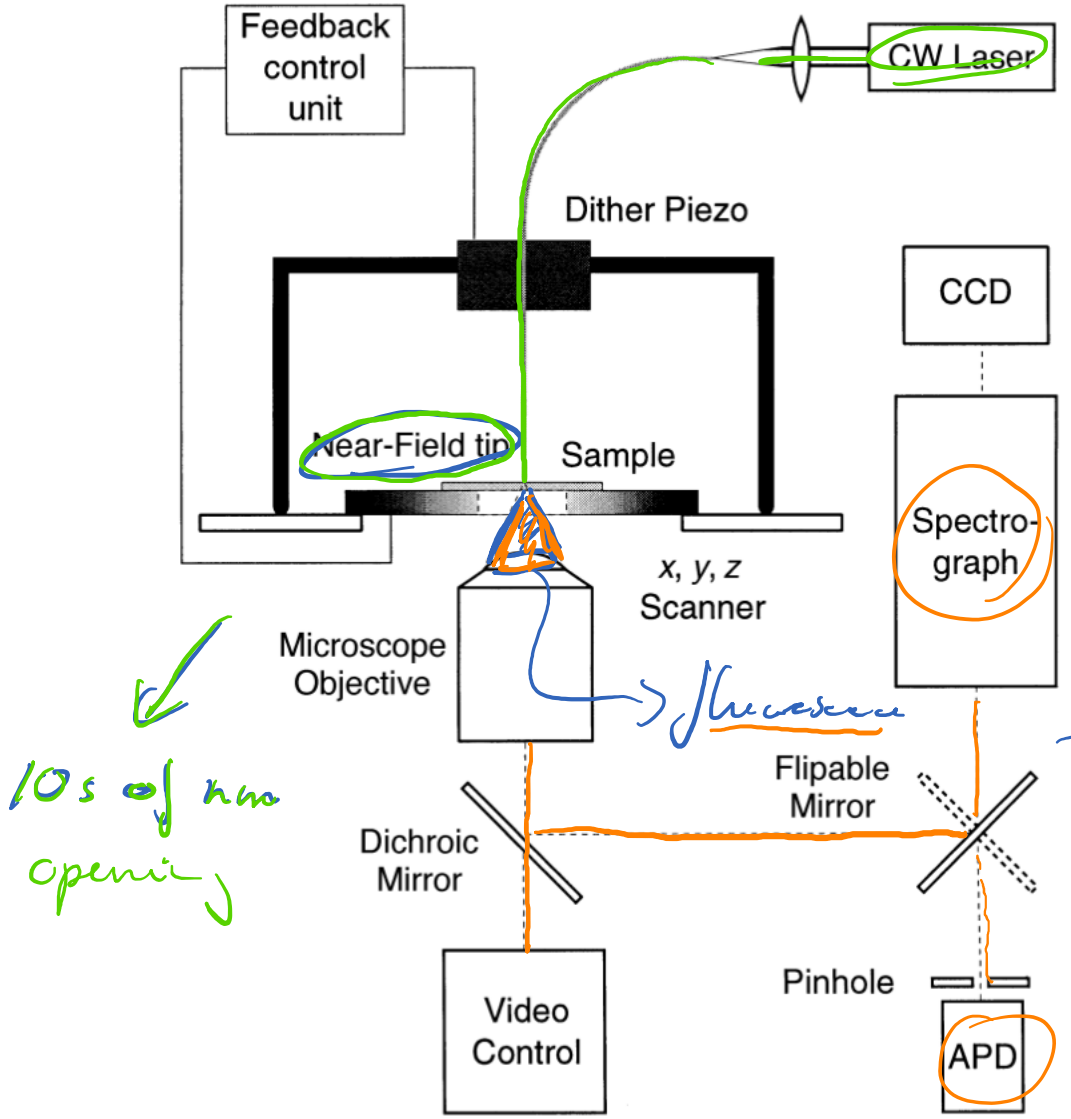
Piezo motors & mechanical stability

(→ SIM / AFM)

SNOM – Scanning Near Field Optical Microscope



Zenobi and Deckert, Angew. Chemie Review, 2000



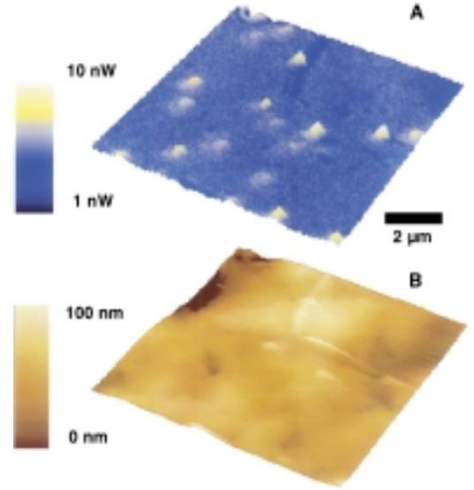


Figure 4. A) SNOM/fluorescence image and B) topographic image of a 10×10 µm area of a PVB film containing fluorescent microspheres. The spheres had a diameter of 288 nm. The fluorescence was excited with the 488 nm line of an Ar-ion laser.

Increasingly fancy approaches

Reviews

A. Hartschuh

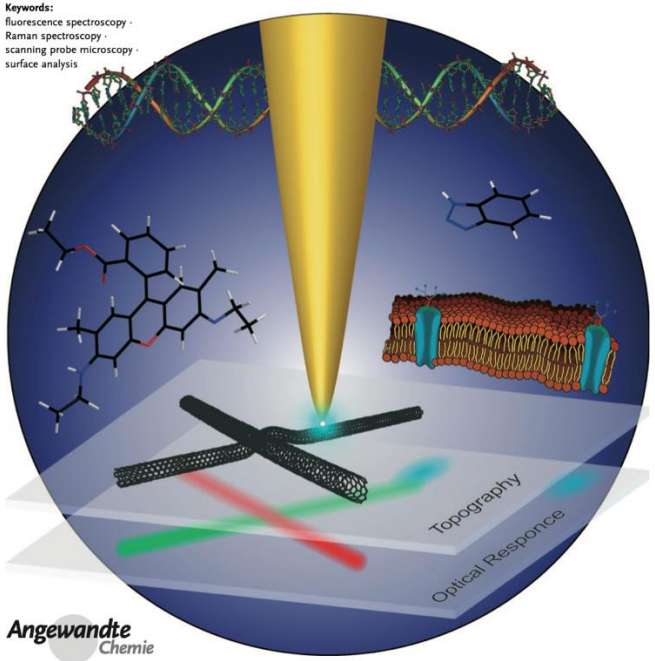
Optics on the Nanoscale

DOI: 10.1002/anie.200801605

Tip-Enhanced Near-Field Optical Microscopy

*Achim Hartschuh**

Keywords:
fluorescence spectroscopy ·
Raman spectroscopy ·
scanning probe microscopy ·
surface analysis



Angewandte Chemie

Angew. Chem. Int. Ed. 2008, 47, 8178 – 8191

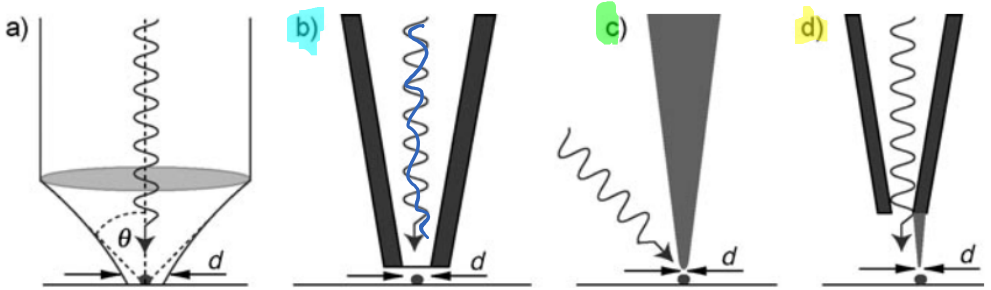


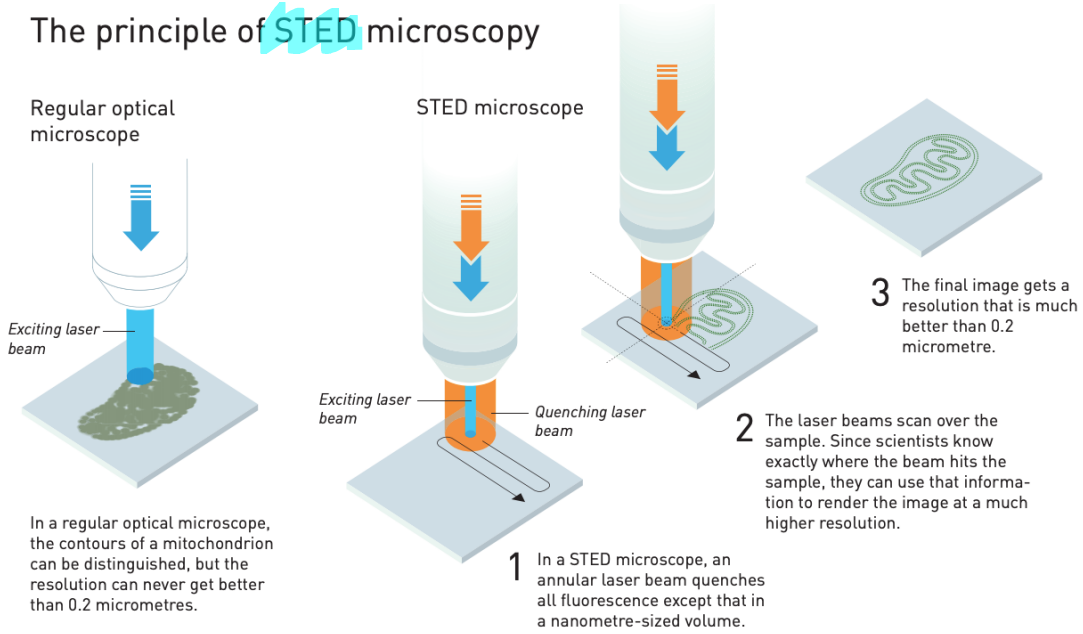
Figure 2. Focusing concepts: a) Far-field focusing using a lens. The angular frequency range of propagating waves $k_{x,max}$, and thus the focus diameter, is limited by the aperture angle of the lens $k_{x,max} = n \sin(\theta) 2\pi/\lambda$, with n being the refractive index and λ the wavelength of light. b) Aperture-type scanning near-field optical microscope (aperture-SNOM). c) Tip-enhanced near-field optical microscopy (TENOM). d) Tip-on-aperture (TOA) approach, which combines the advantages of (b) and (c).

Hartschuh, Ang. Chemie, Review 2008

Limitations: – need to be close to surface
– not so good for large or living samples

Last comment: Remember session 1?

Recent examples of applications of light to chemistry



The Nobel Prize in Chemistry 2014

© Nobel Media AB. Photo: A. Mahmoud

Eric Betzig

Prize share: 1/3

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Stefan W. Hell

Prize share: 1/3

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William E. Moerner

Prize share: 1/3

→ fluorescence microscope

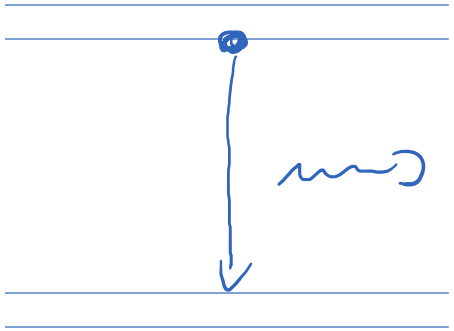
→ scanning

→ other tricks

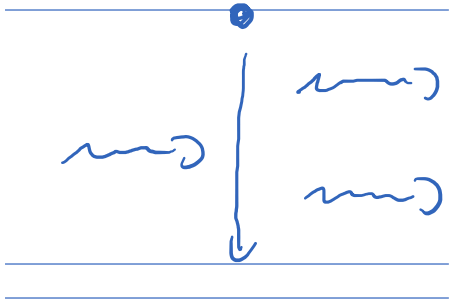
Eric Betzig, Stefan W. Hell *and* ***William E. Moerner*** *are awarded the Nobel Prize in Chemistry 2014 for having bypassed a presumed scientific limitation stipulating that an optical microscope can never yield a resolution better than 0.2 micrometres. Using the fluorescence of molecules, scientists can now monitor the interplay between individual molecules inside cells; they can observe disease-related proteins aggregate and they can track cell division at the nanolevel.*

From Lecture 5:
Absorption, emission, stimulated emission - overview

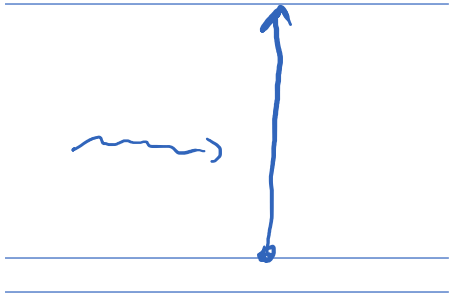
Spontaneous emission



Stimulated Emission



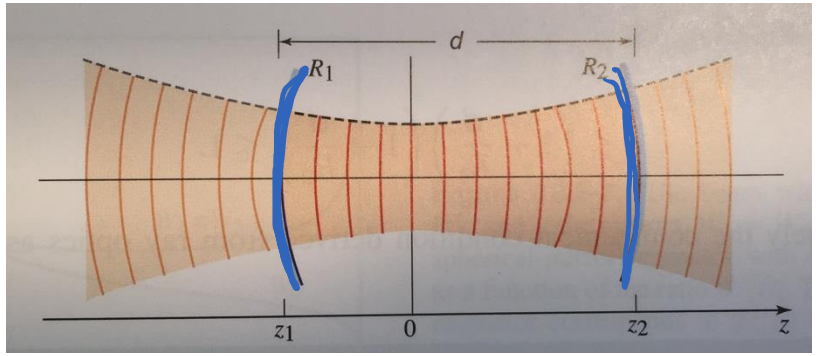
(Stimulated) absorption

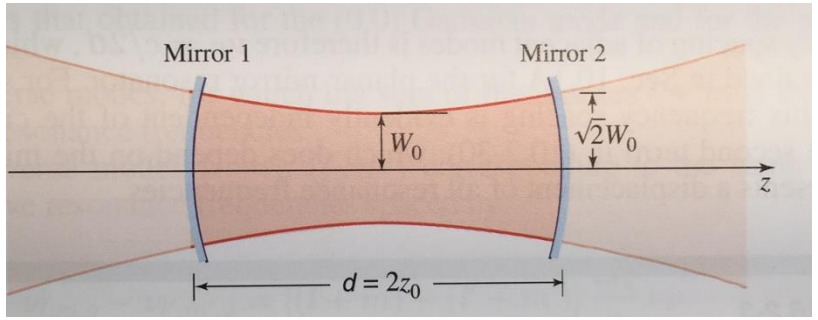


absorption
⇒ fluorescence
⇓
fluorescence microscope

Implication: ⇒ if you come in with enough light, you can "deplete" levels
⇒ balance between absorption & emission
⇒ high intensities can suppress fluorescence

From Lecture 4:
Gaussian Beam Resonator

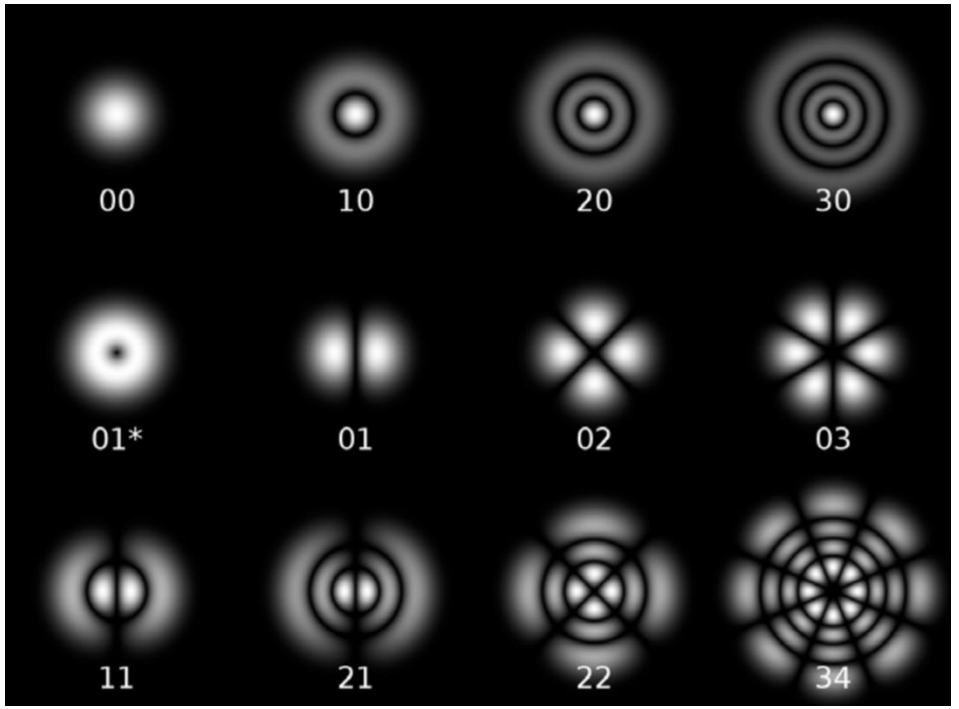


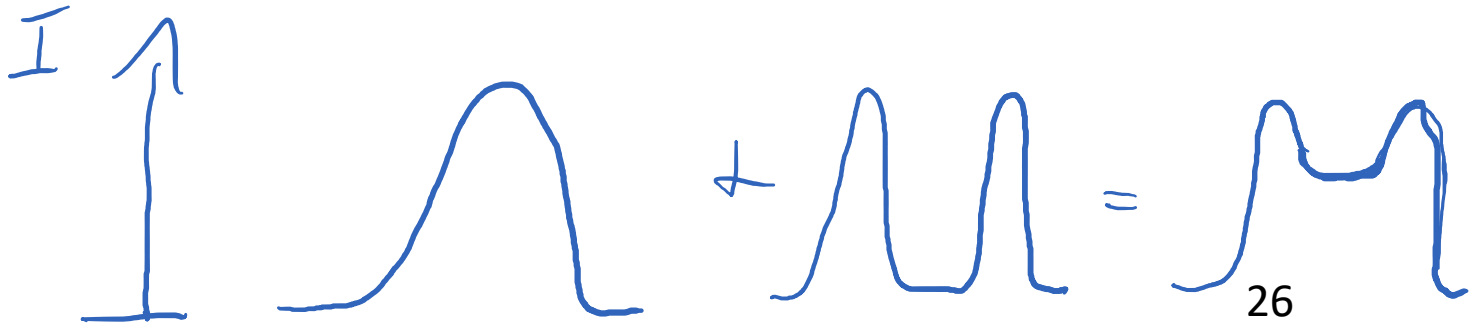


Laguerre-Gaussian modes

Gaussian

"Donut"



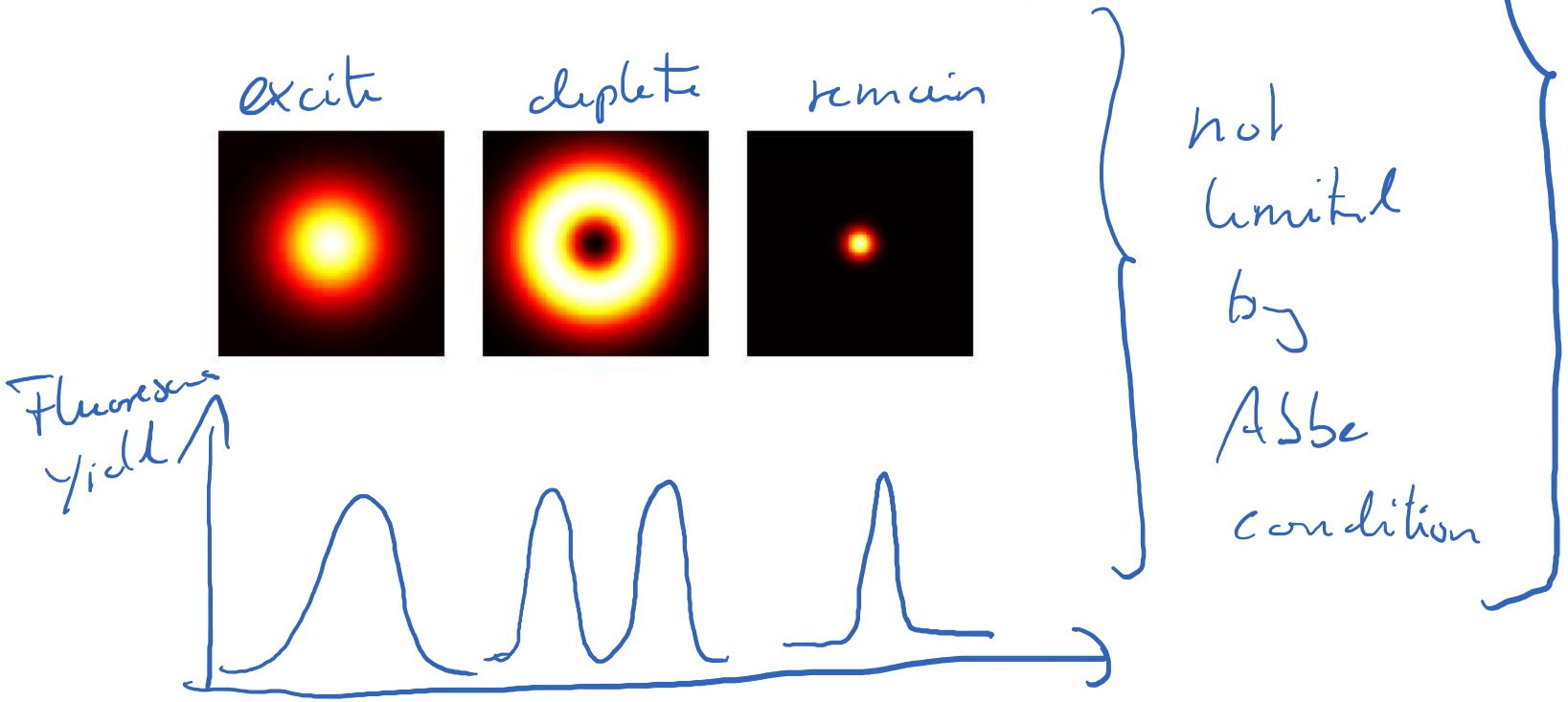


Stefan Hell: Stimulated Emission Depletion Microscope (STED)

- Based on Fluorescence Microscope
- Based on Laser Scanning Microscope

But with Important additional concepts:

Remember Gaussian, Bessel beams, focal spots



Enjoy video: https://www.youtube.com/watch?time_continue=428&v=0NCNy6pVIZE

The end.