Microscopy: Principles and Advances

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Academic Background

2005-2008: PhD-Chemical Biology

Prof. Richard Templer, Dr. Oscar Ces, Prof. John Seddon (Chemistry) Prof. So Iwata (Biochemistry), United Kingdom

Imperial College London

MARIE CURIE ACTIONS

2008-2010: Postdoctoral Research Assistant

Prof. Otto Glatter, Scattering Methods Research Group
Chemistry-University of Graz, Austria

UNI GRAZ Scattering Methods

2011-2011: Postdoctoral Research Associate

Prof. Matthias Weiss, Experimental Physics I
Physics-University of Bayreuth, Germany

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2012-2013: Postdoctoral Research Associate

Dr. Ulrich F. Keyser, Nanopores Research Group
Physics-University of Cambridge, United Kingdom

UNIVERSITY OF CAMBRIDGE

2013-……: Assistant Professor and Group Leader

Lipid Nanostructures Group, Centre For Materials Science
University of Central Lancashire, United Kingdom

uclan University of Central Lancashire

Microscopic Eye!

Can you see it with eyes?

Can you make it seeing with eyes?

Why to Use Microscopes?

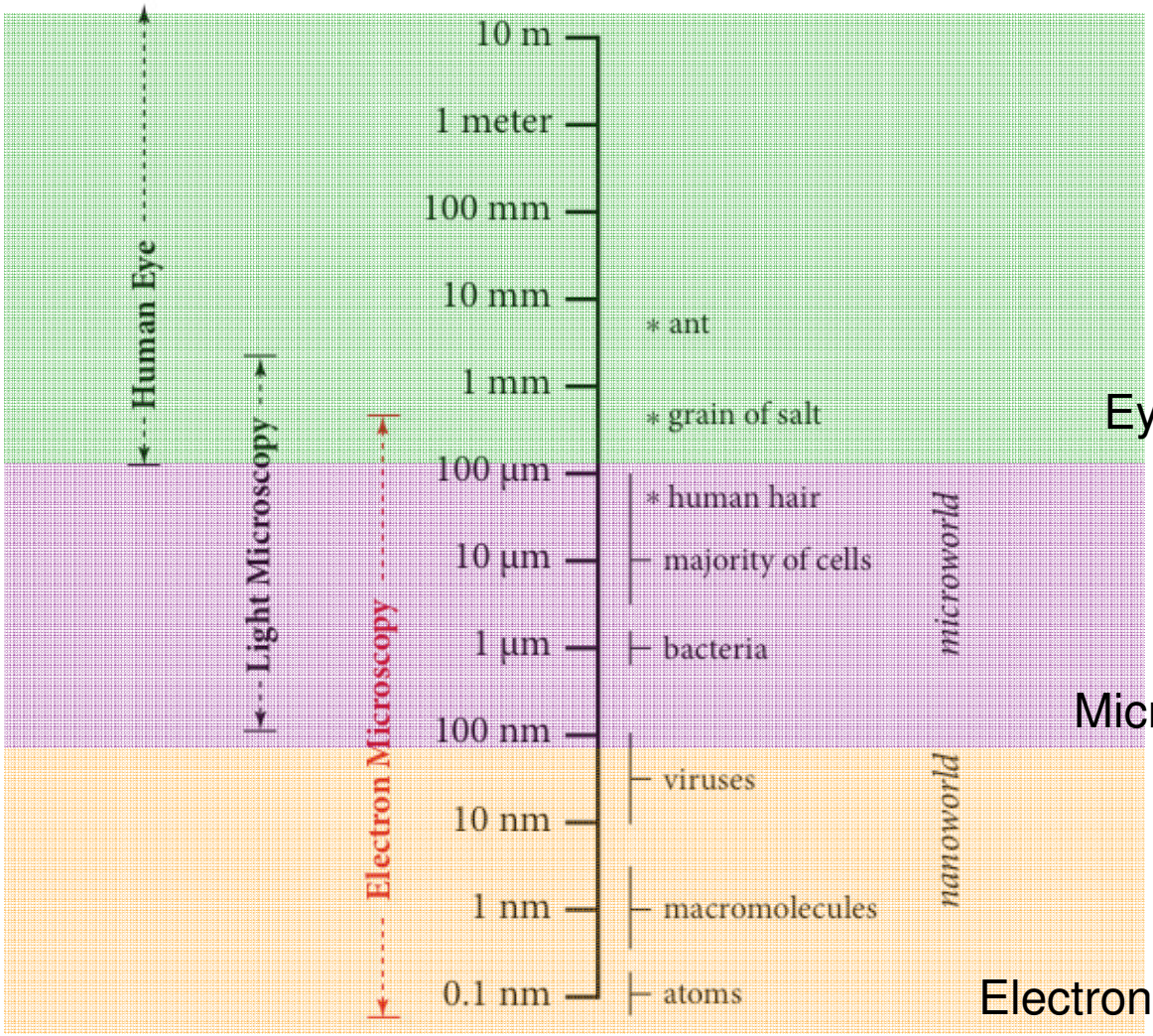
1. See things (objects,orgamisms) that are not visible with naked eye

2. Study morphological properties at micro- and nano-scale lengths

3. Collect images at high resolution

4. Observe live phenomena (live cells, chemical reaction) under microscope

Detection Limits



Eye: 0.1 mm

Human hair

Blood Cells

Microscope: x1000

DNA, organelles

Electron Microscope: x1000x1000

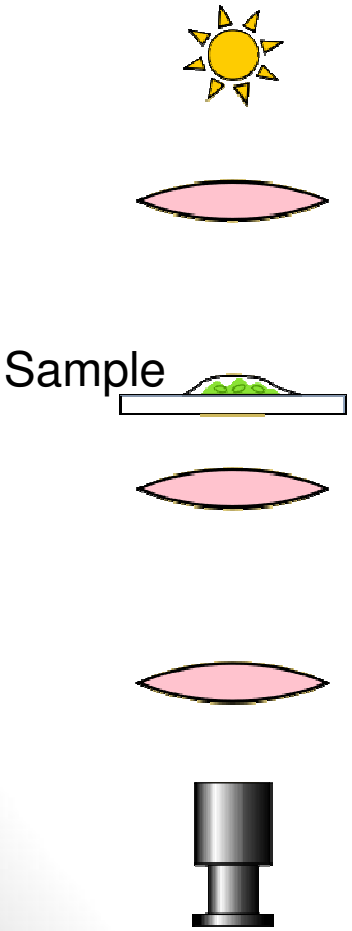
Types of Microscopes

Types based on what interacts with sample

Light

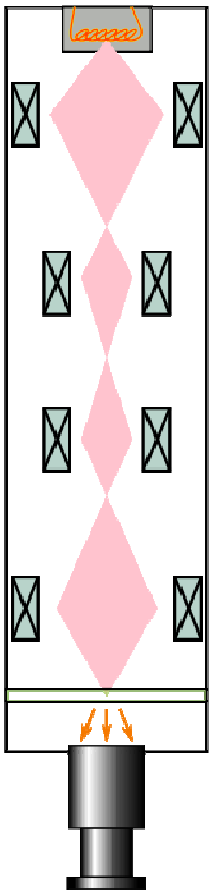
Optical

Sample

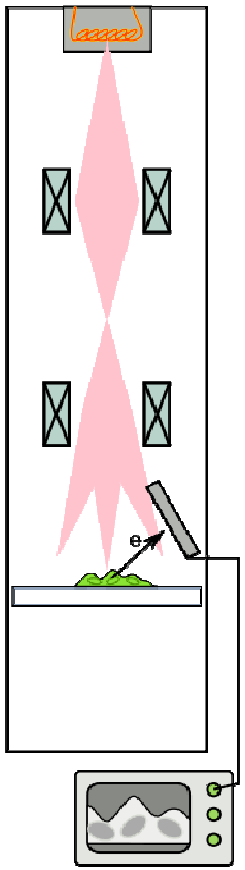


Electrons

TEM



SEM



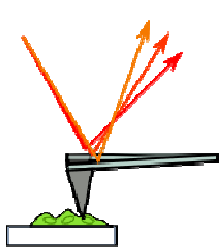
Probe

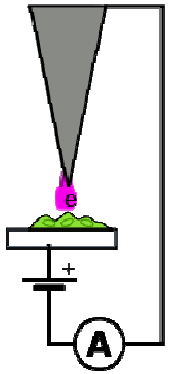
SPM

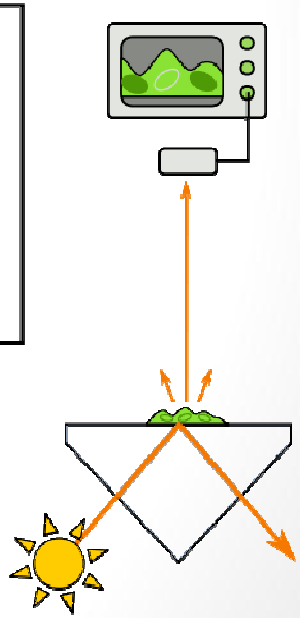
AFM STM SNOM

scanning tunneling microscopy

scanning near-field optical microscopy







Light Microscopes

Optical

Light source

Lens: to focus light on sample

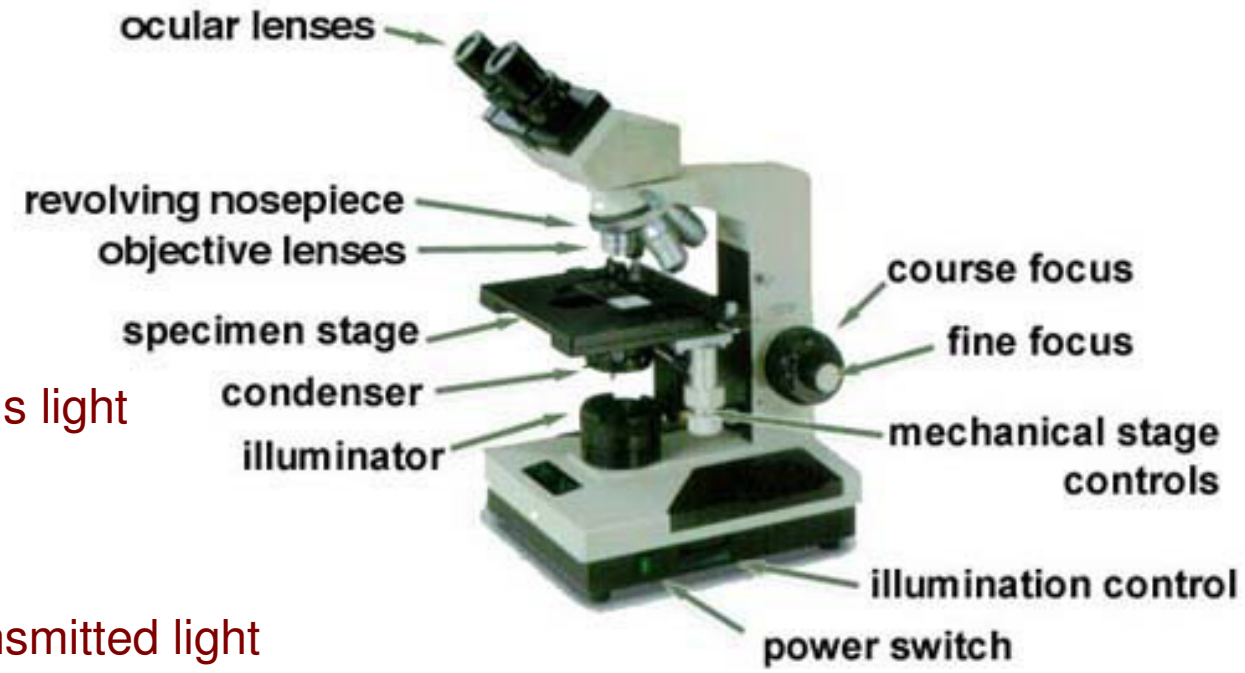
Sample

Transmitted light

Stacking of lenses to increase magnification

Lens: to focus light into an eye, objective, camera

eye, objective, camera



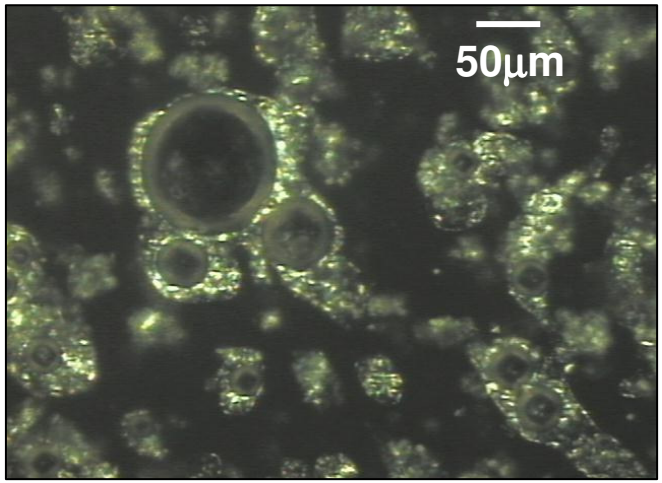
Further Types of Light Microscopes

- Bright Field: simplest, used for basic observations
- Dark Field: better contrast but reduces light illumination
- Polarized Light: anisotropic samples, based on birefringence
- Phase contrast: uses difference in refractive indices

Shrikhand with Nuts, Saffron

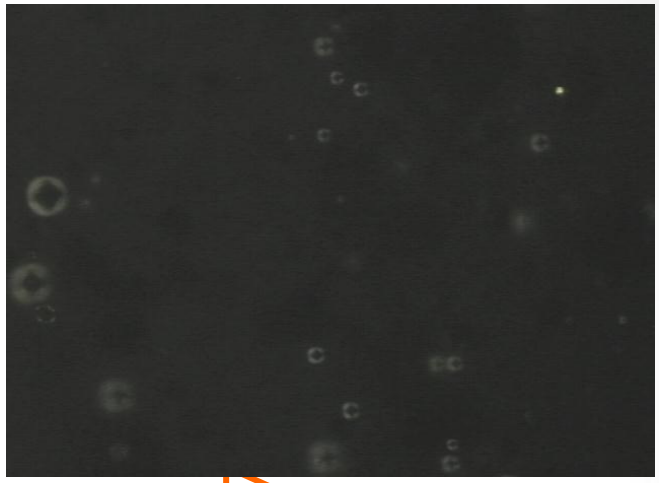
Melting and Re-crystallization of Lipids/Fats

Liquid Crystalline Fat Structures

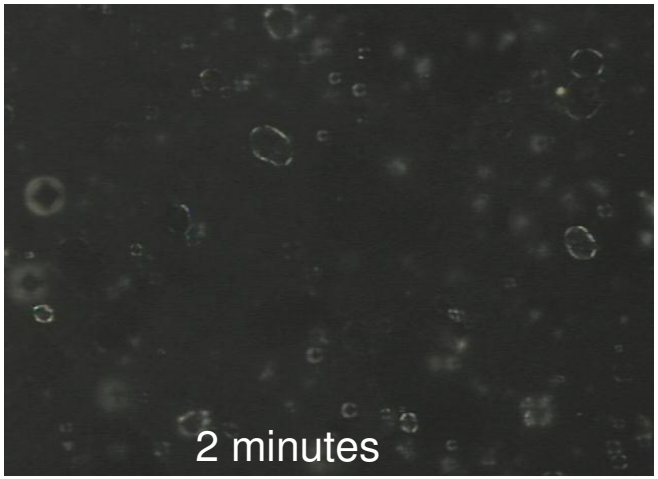


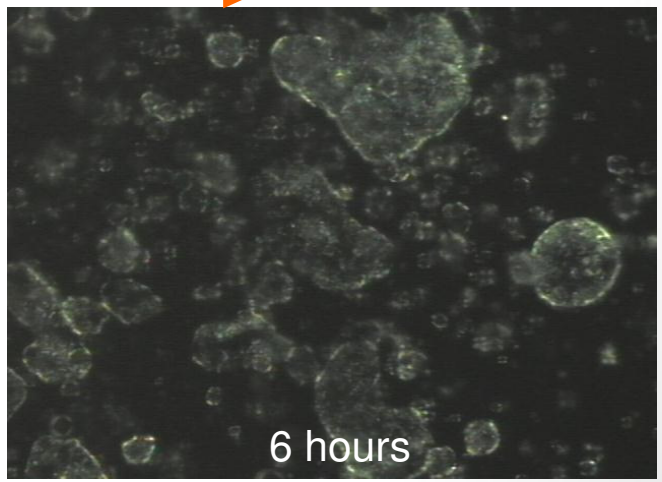
Annealing at 40°C

Melting of Fat Structures



Fat Re-crystallization at Room Temp. (cross polarized light)

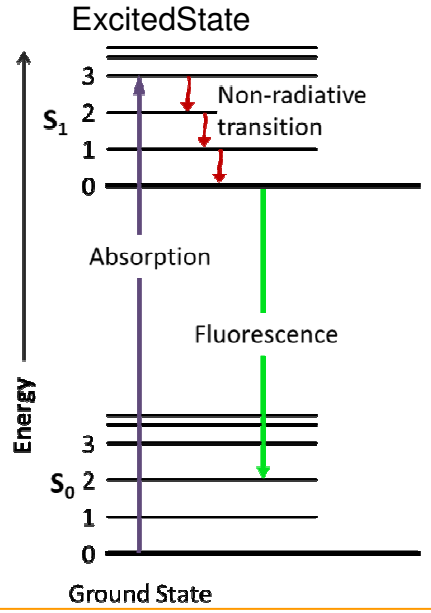


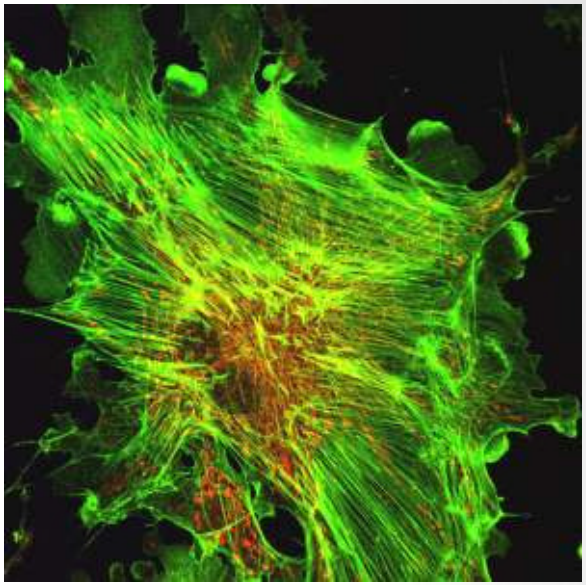


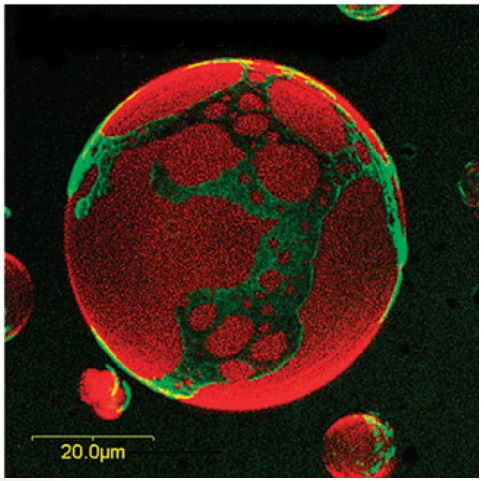
Fluorescence Microscopy

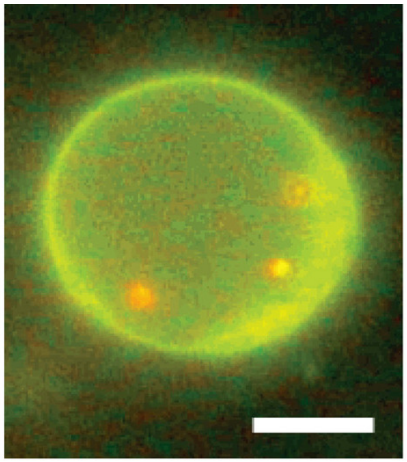
Fluorescent Microscopy

Excite with high energy light, they emit light of a different, lower frequency (long wavelength)

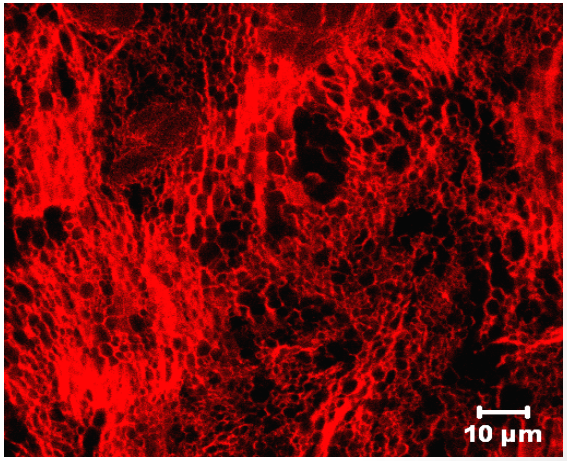






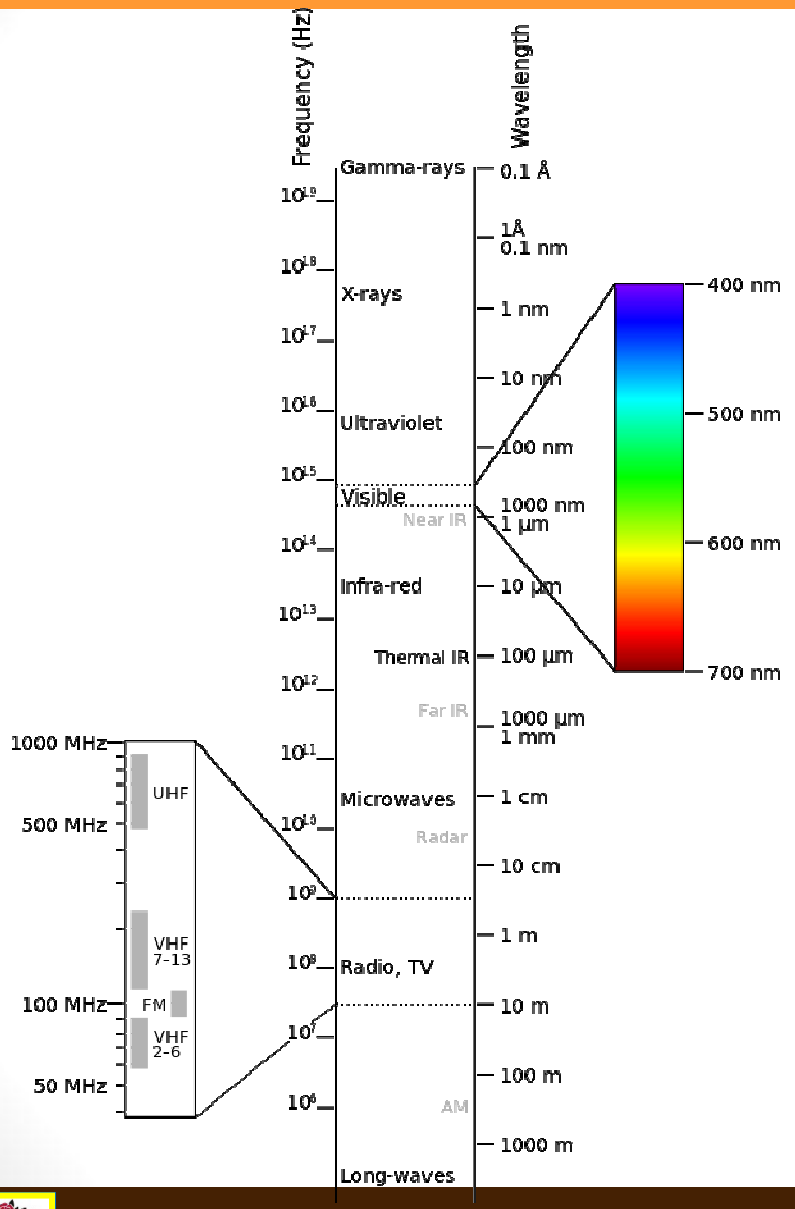


Giant Unilamellar Vesicles



W/O nanostructured emulsion

Other than Visible Light Microscopy



X-ray Microscope

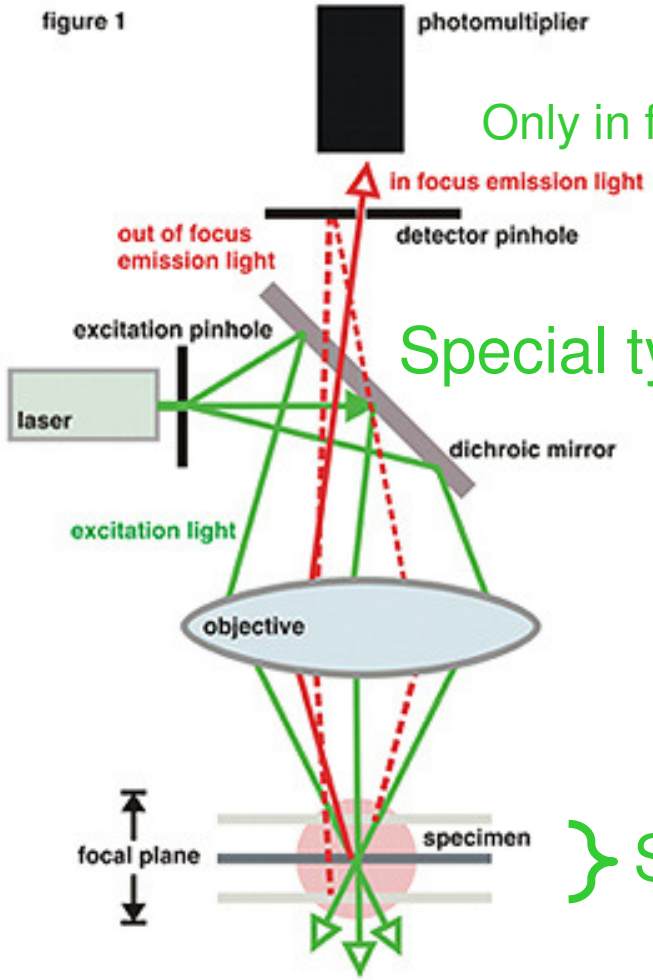
UV Microscope

Visible Light Microscope

IR Microscope/Raman Microscope

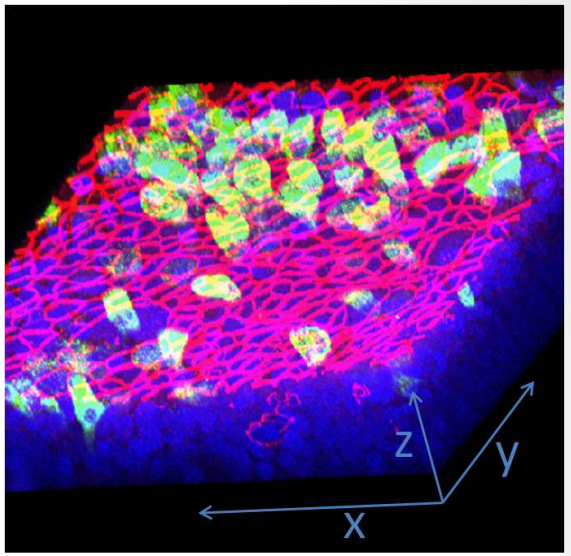
Confocal Microscopy

Limitations of wide field microscopes:
Low resolution, Out of focus light, Diffracted light



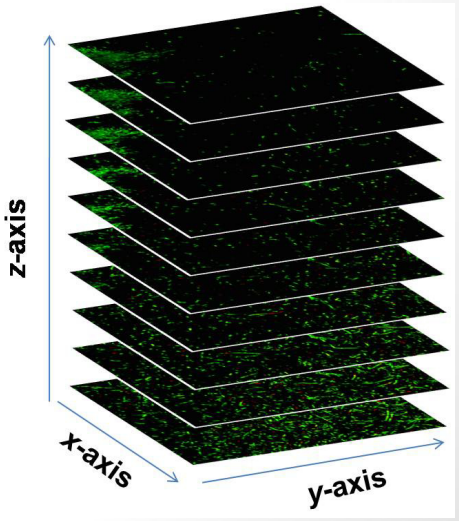
Only in focus light is detected

Special type of mirror



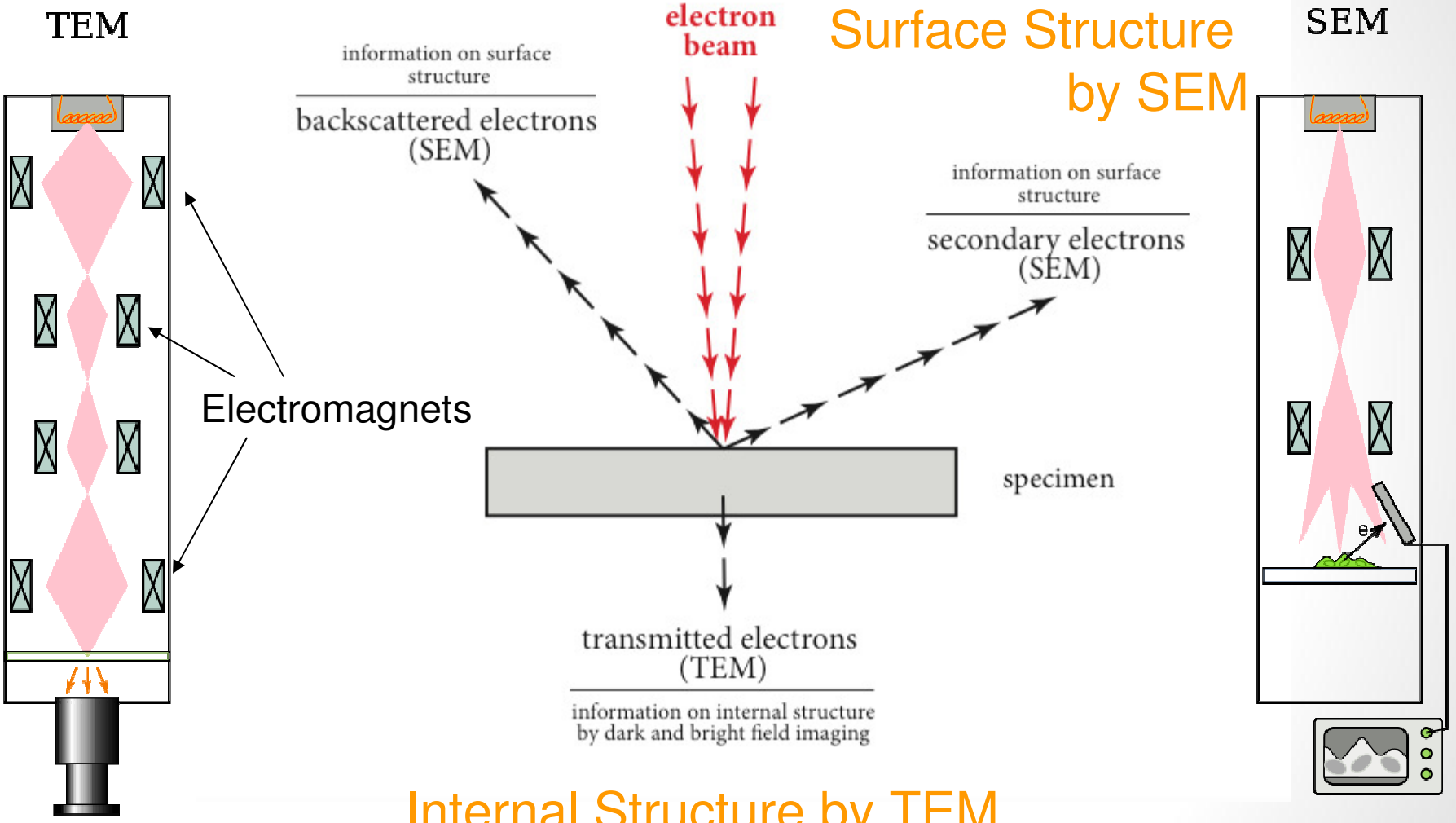
3-D image reconstruction

Z-stacking



} Study plane

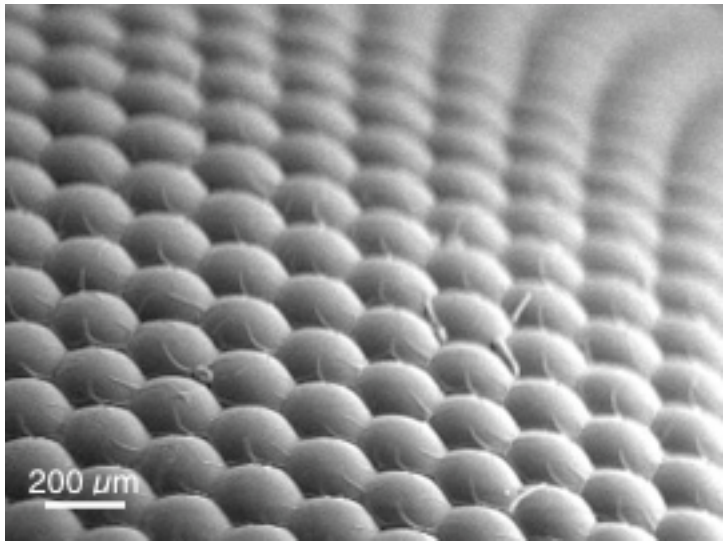
Electron Microscopy



Surface Structure by SEM

Internal Structure by TEM

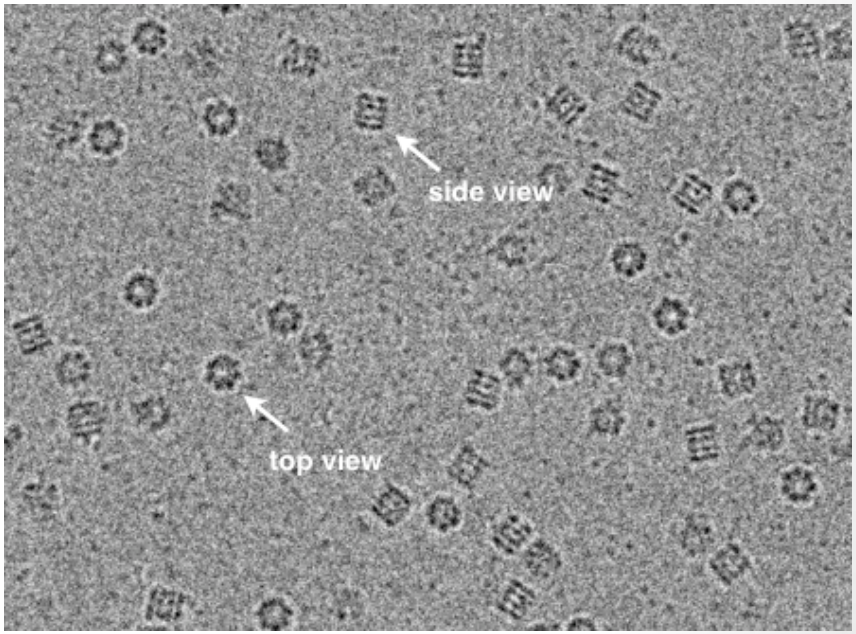
Examples: Electron Microscopy



SEM

Eye surface of housefly

TEM

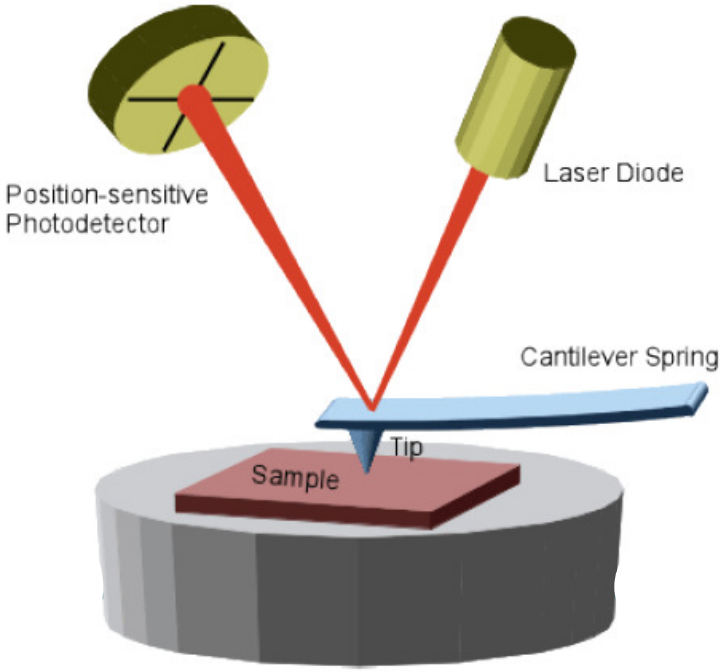


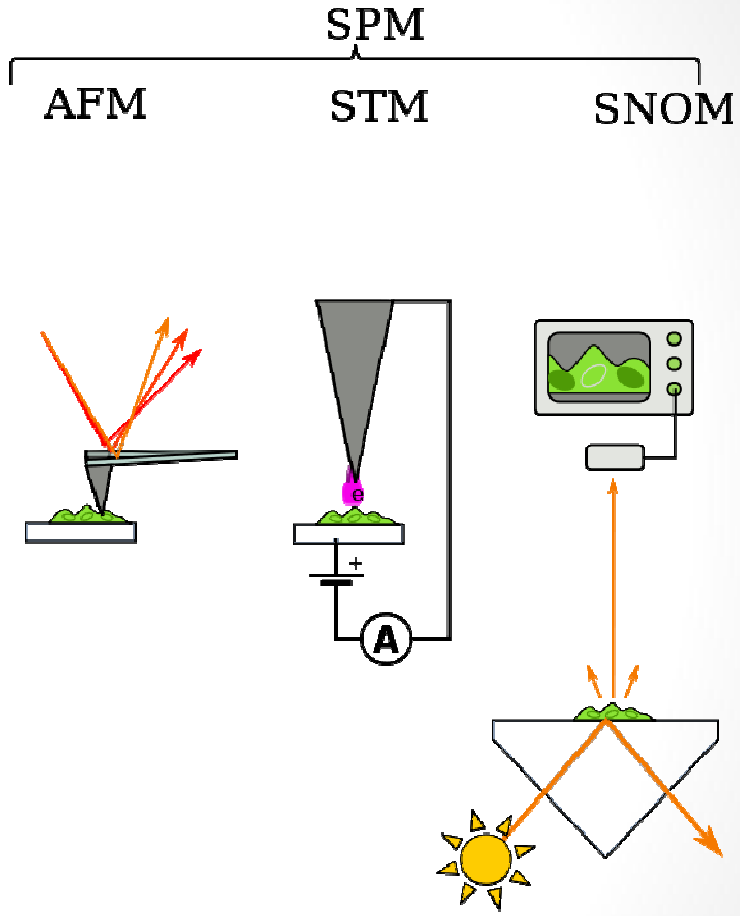
Cryo-to keep structure intact
(biological and soft samples)

Cryo-EM image of GroEL at 50,000X magnification.
The GroEL molecular machine that functions in folding of many proteins in cells.

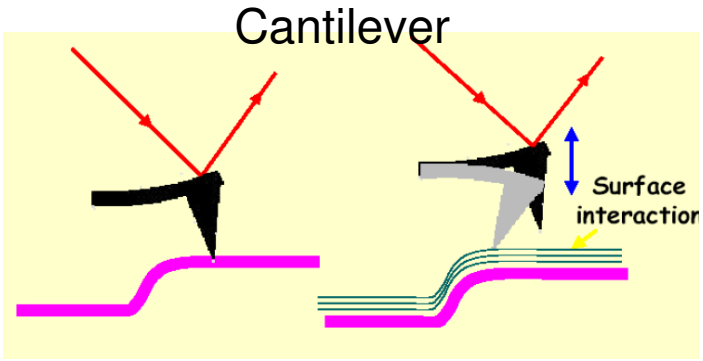
Probe Microscopy

AFM:
Atomic Force Microscopy

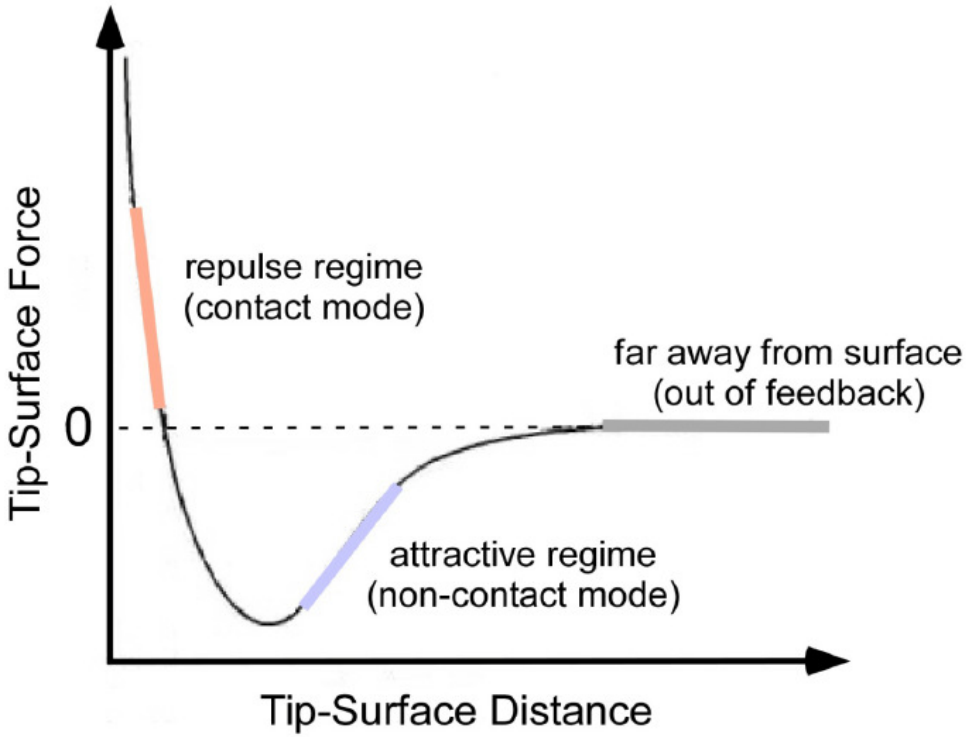




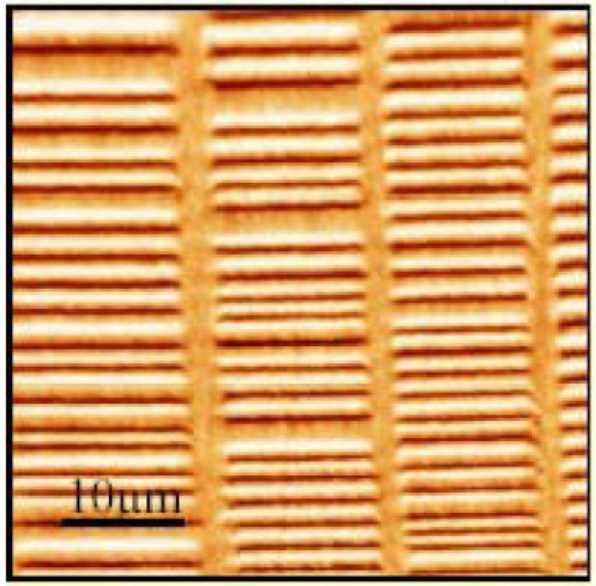
Modes of Cantilever



Contact mode Non-contact mode

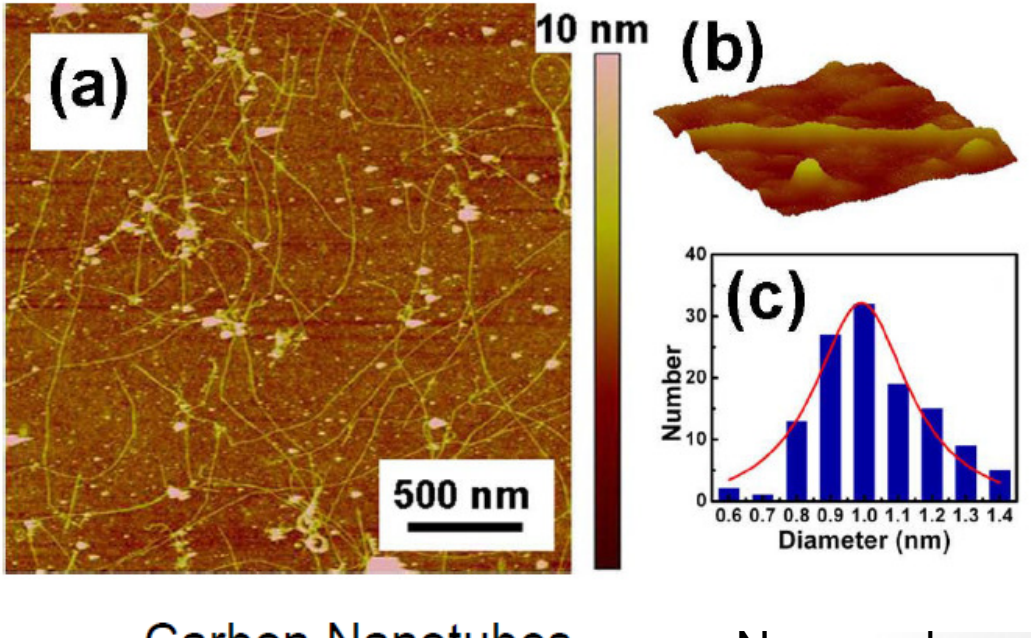


Images by Atomic Force Microscopy



Magnetic bits of a zip disk

Surface topography



Carbon Nanotubes

Nanoscale dimensions

Micro- to nano-scale information

Advances in Microscopy

Techniques with Confocal Laser Scanning Microscopy:

- Fluorescence Correlation Spectroscopy (FCS)
- Live Cell Imaging
- Fluorescence Lifetime Imaging Microscopy (FLIM)
- Forster Resonance energy Transfer (FRET)
- Fluorescence Recovery After Photo-bleaching (FRAP)

Super-resolution Microscopy

Total Internal Reflection Microscopy (reaching nano by an eye)

Mutiphoton Microscopy

Contact Details

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Thank You!