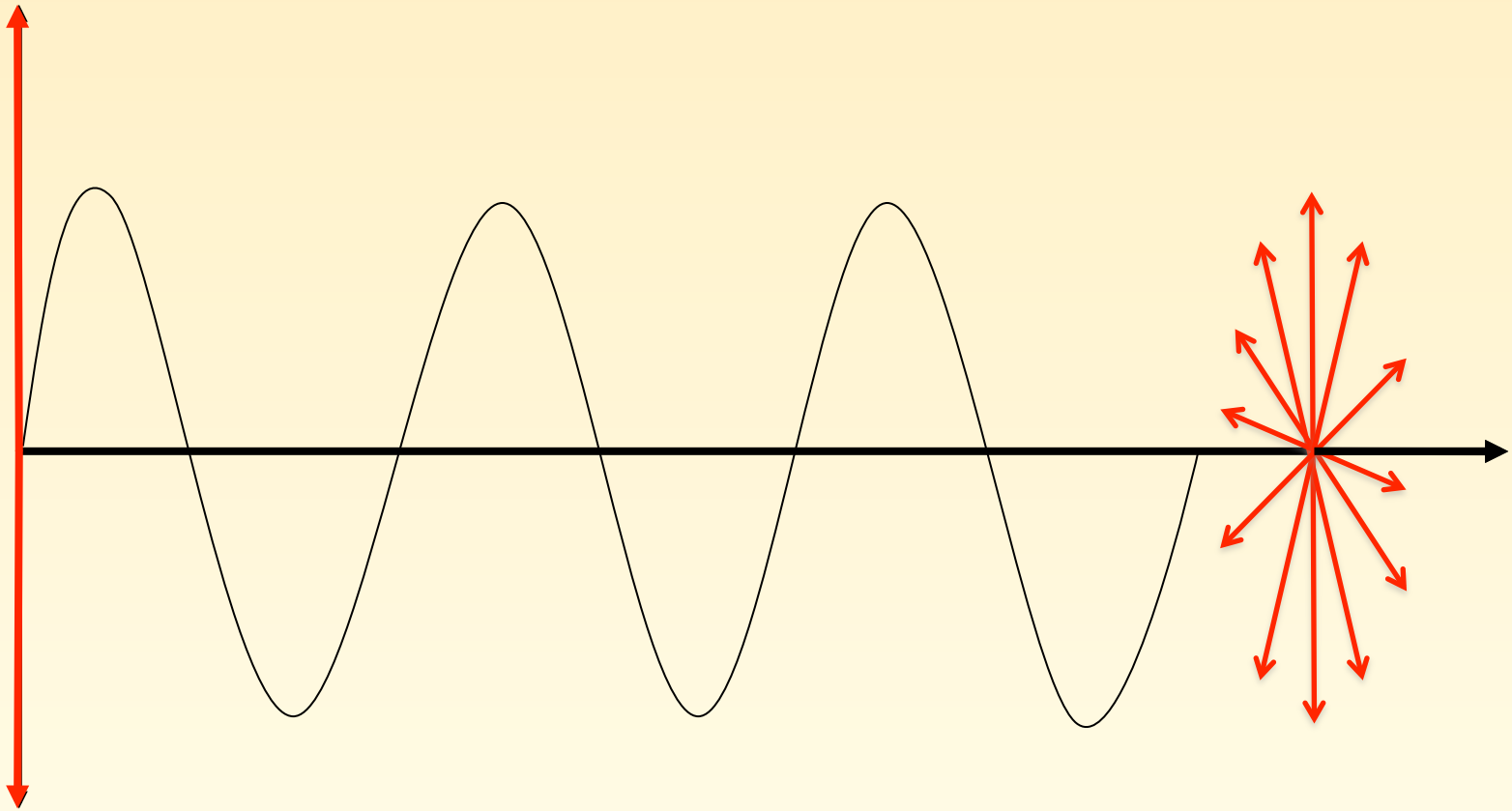
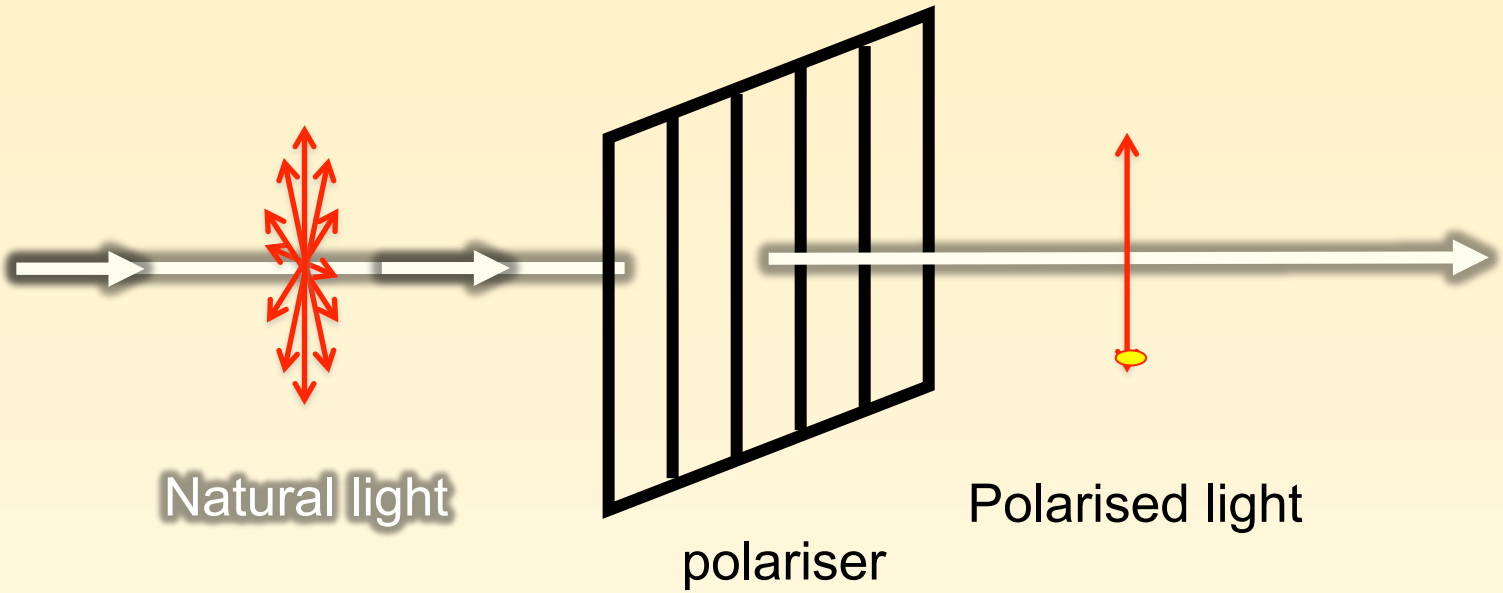
4. Polarized Light



Light has properties of particles and waves. If considered as a wave the planes of vibration are perpendicular to the propagation direction.

In ‘natural light’ these vibrations occur in all directions – nonpolarized light.

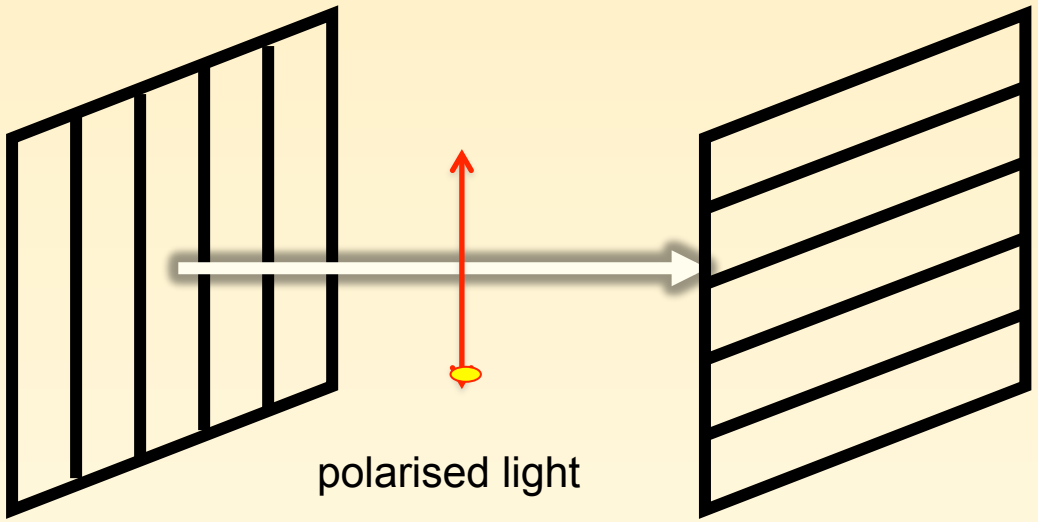
4. Polarized Light



Natural light vibrates in all directions

In **Polarised Light** all but one of these directions have been ‘filtered out’.

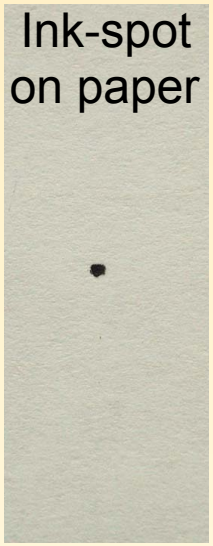
4. Polarized Light

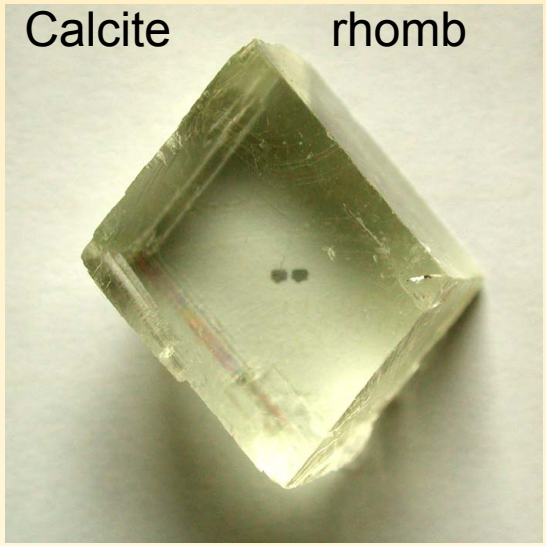


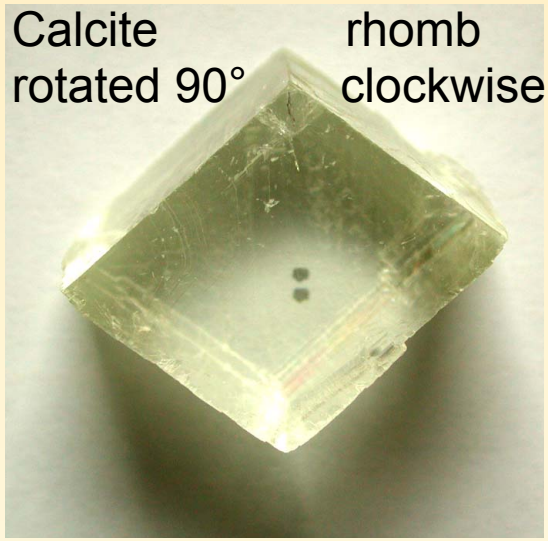
NO light
transmitted

Crossed Polars → **Darkness**

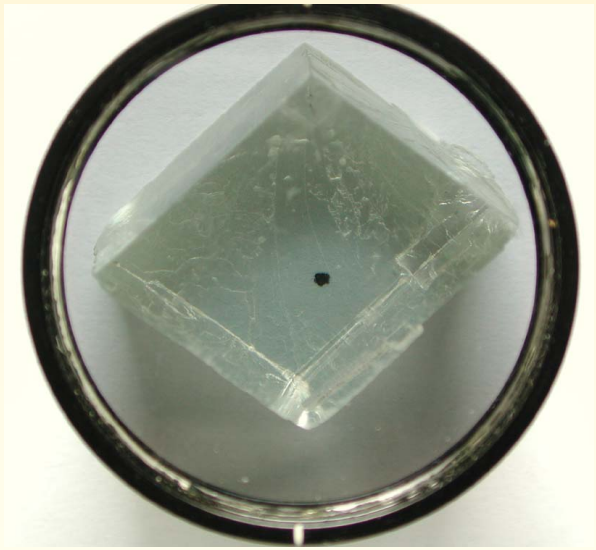
4. Polarised Light - Birefringence

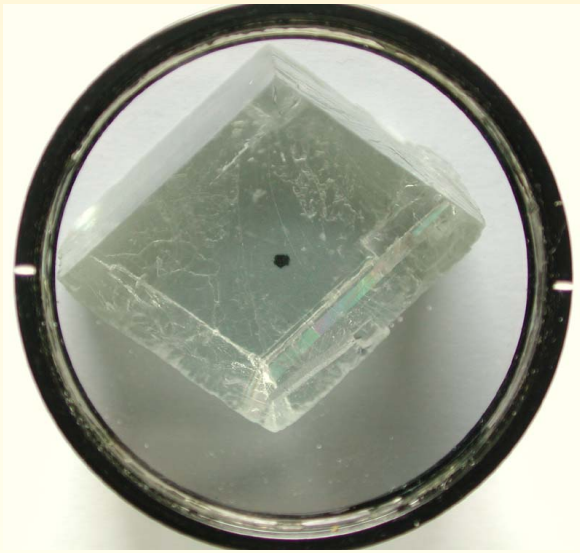




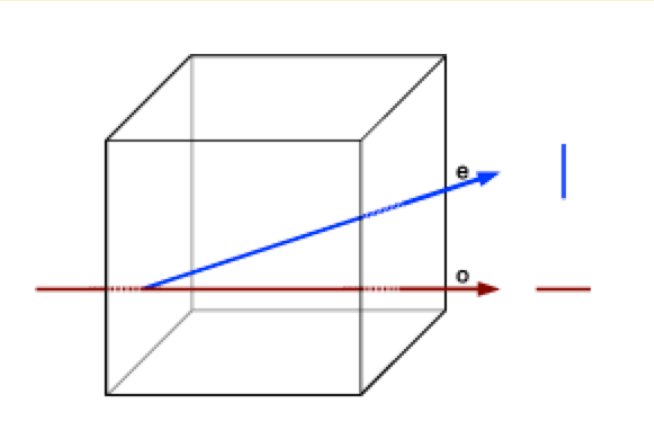


Double image of spot appearing in different xyz positions





4. Polarised Light - Birefringence

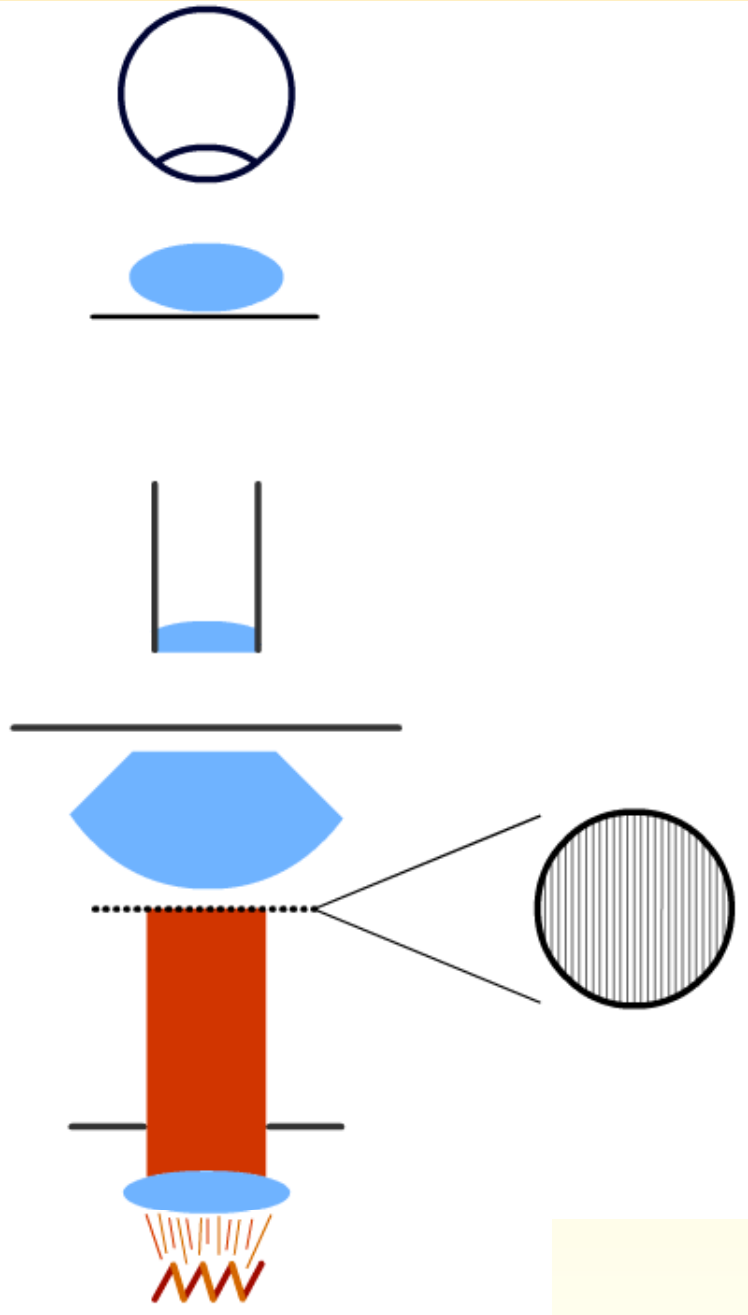


Birefringent / anisotropic materials:
- Separate a beam of light into two beams or waves
- Each wave is linearly polarized
- Polarisation direction perpendicular
- Refractive index depends on propagation direction

Ordinary beam (o):
- no refraction when incidence angle 90°

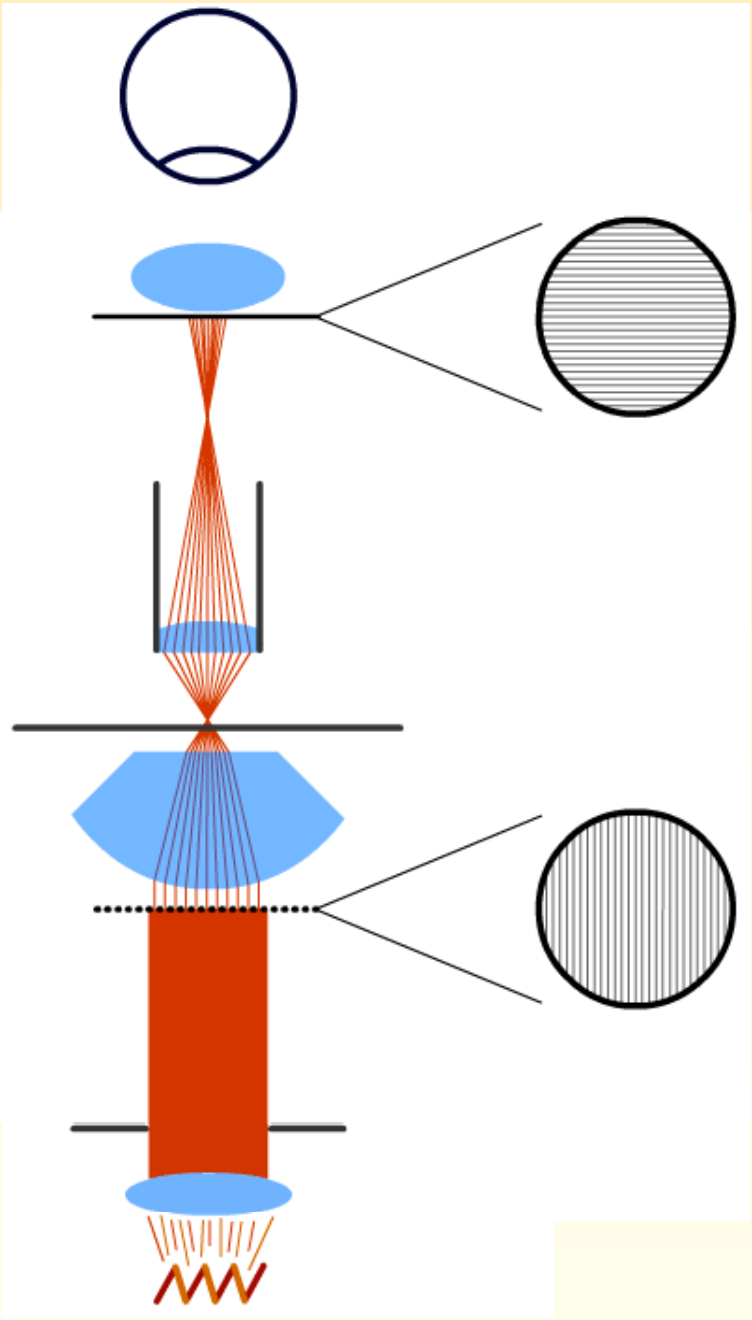
Extraordinary beam (e):
- refractive index depends on incidence angle
  → refraction of the beam

Different speed of **o** & **e** due to Δn → phase shift when leaving material



Polariser

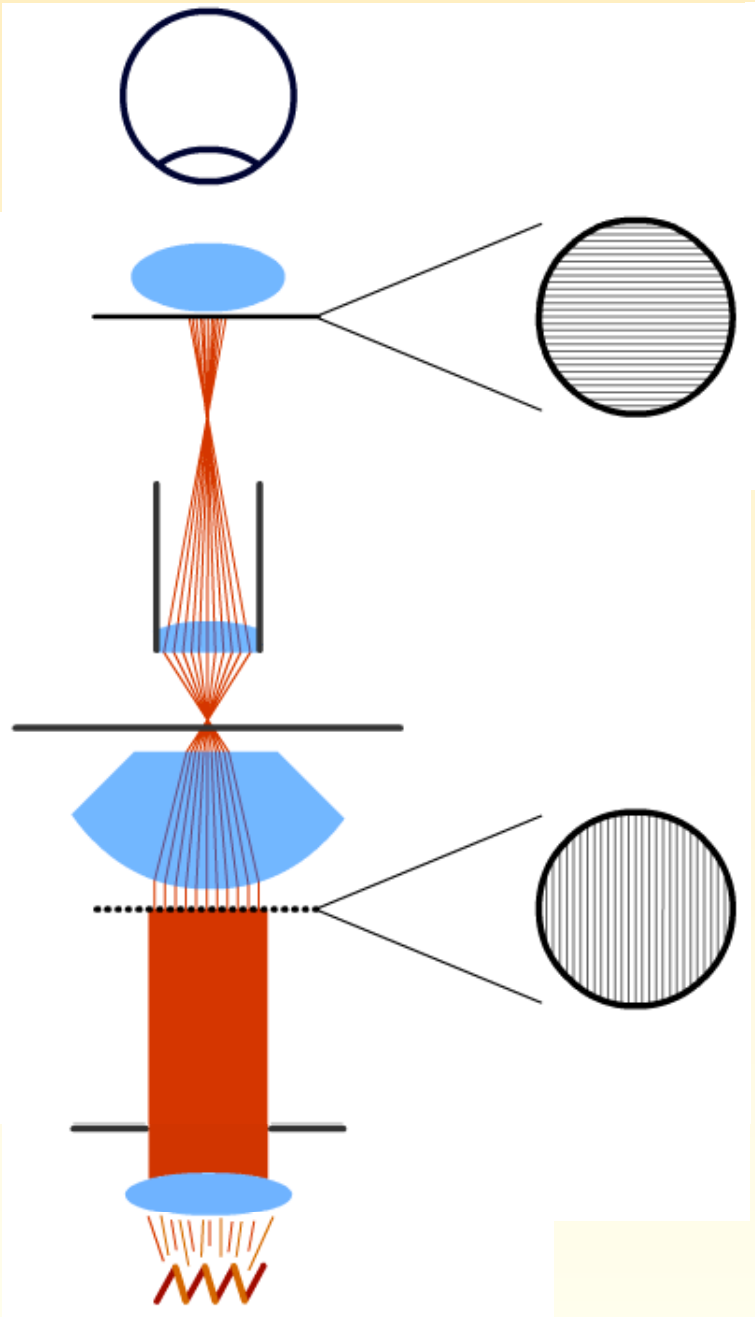
See: http://www.univie.ac.at/mikroskopie



Analyser
perdendicular to polariser

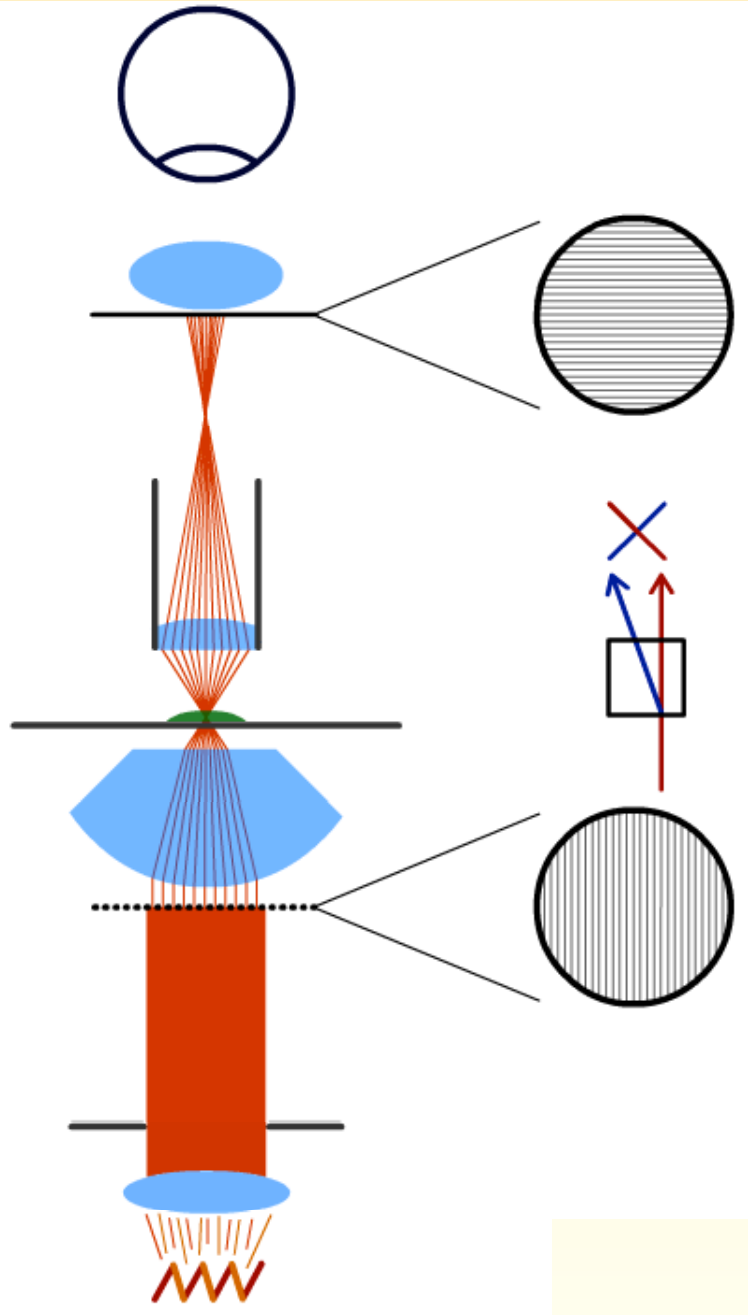
Polariser

See: http://www.univie.ac.at/mikroskopie



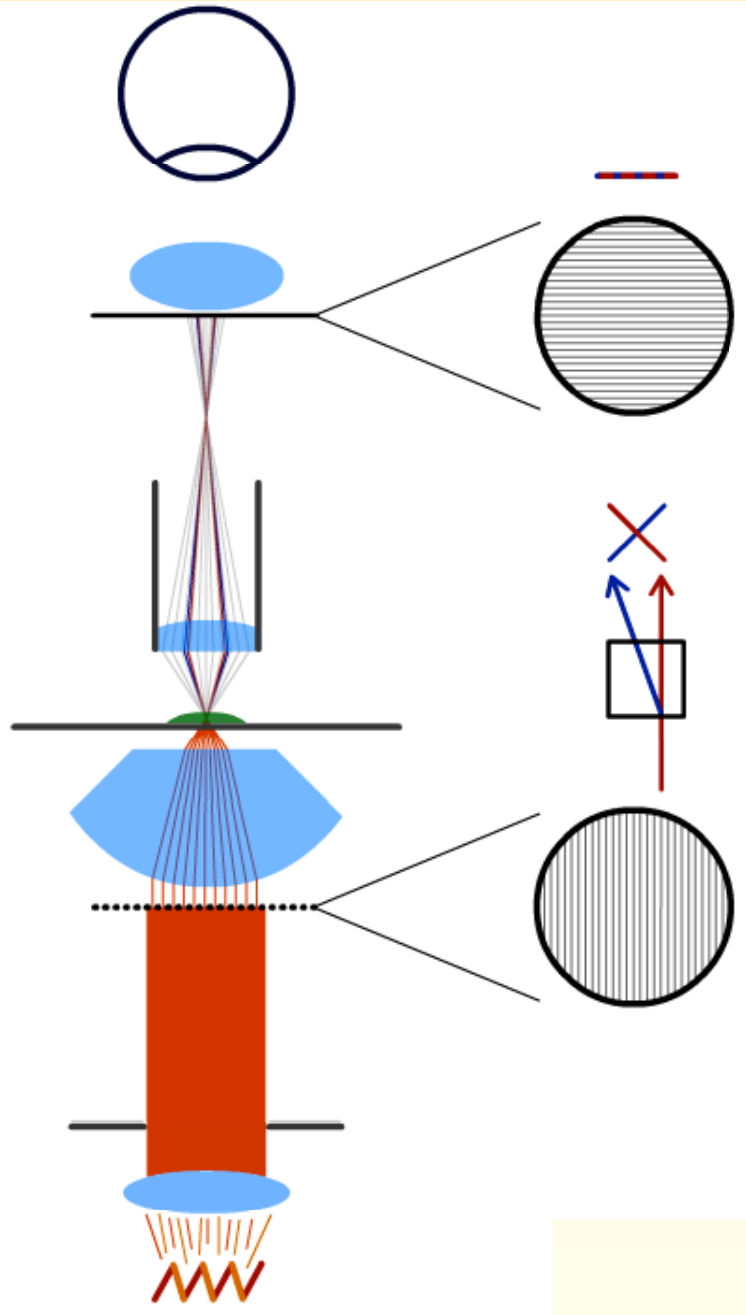
No object, crossed polars

See: http://www.univie.ac.at/mikroskopie



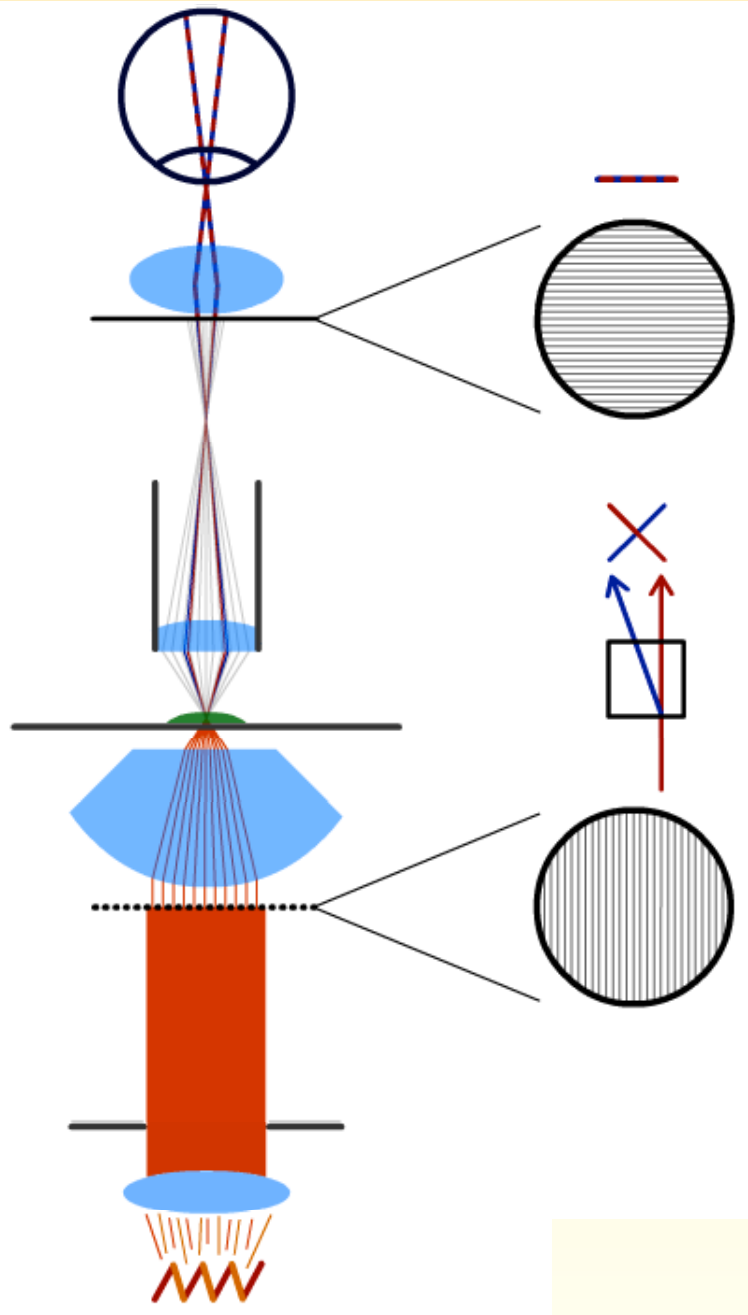
Birefringent object
waves are split into o and e

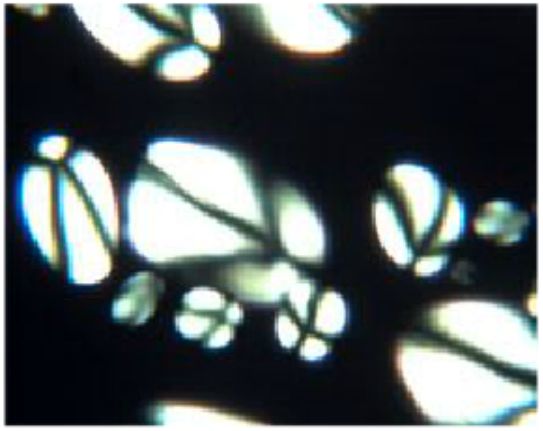
See: http://www.univie.ac.at/mikroskopie



Interference at analyser
because polarisation direction rotated to the same plane (like analyser)
loss of intensity

See: http://www.univie.ac.at/mikroskopie

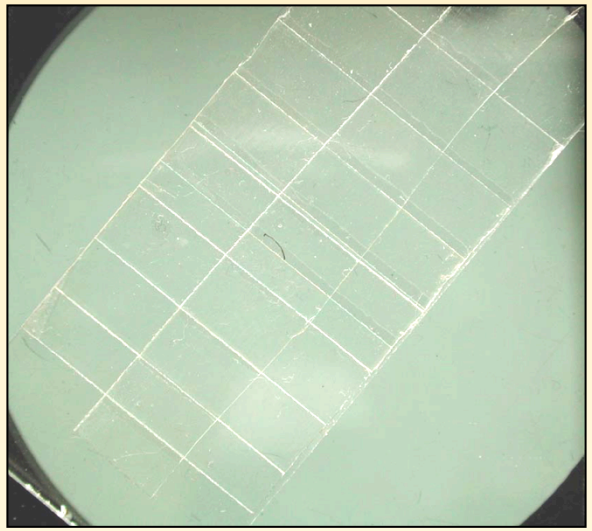




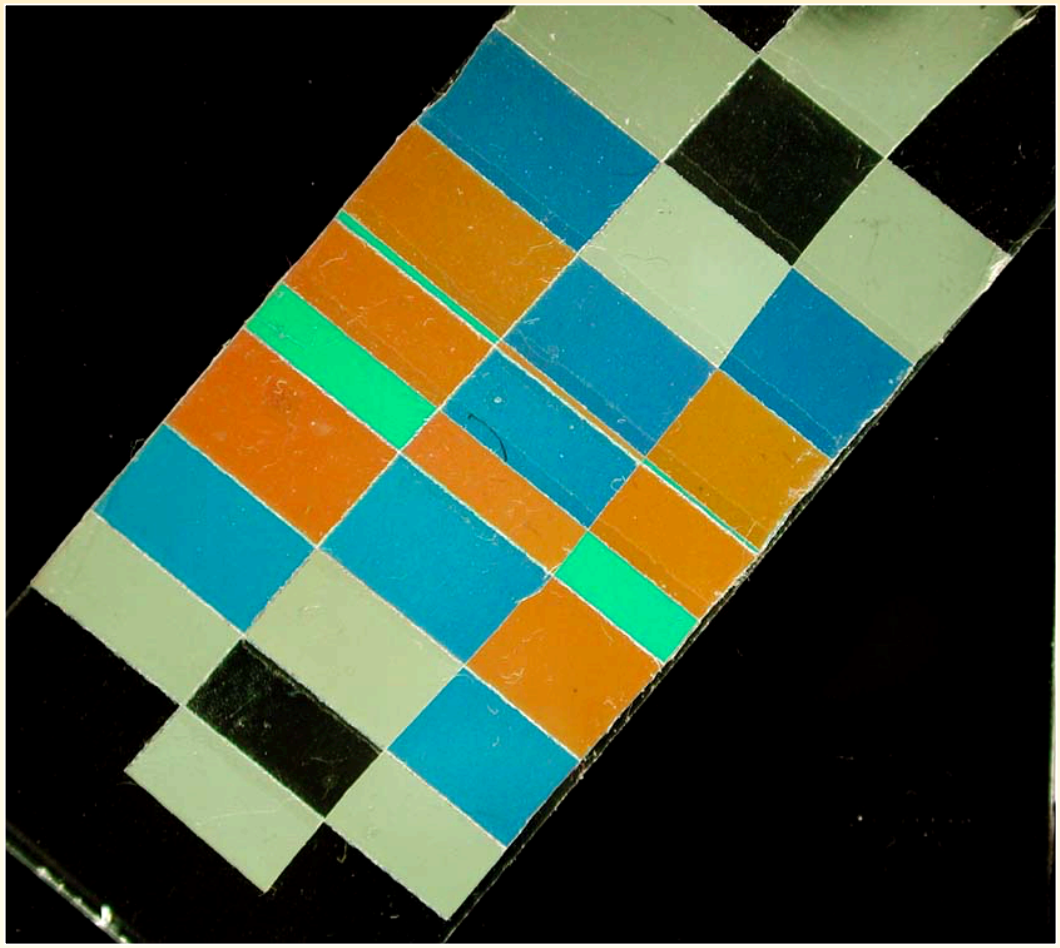
Constructive Interference
Lights up birefringent structures

See: http://www.univie.ac.at/mikroskopie

4. Polarised Light - Example



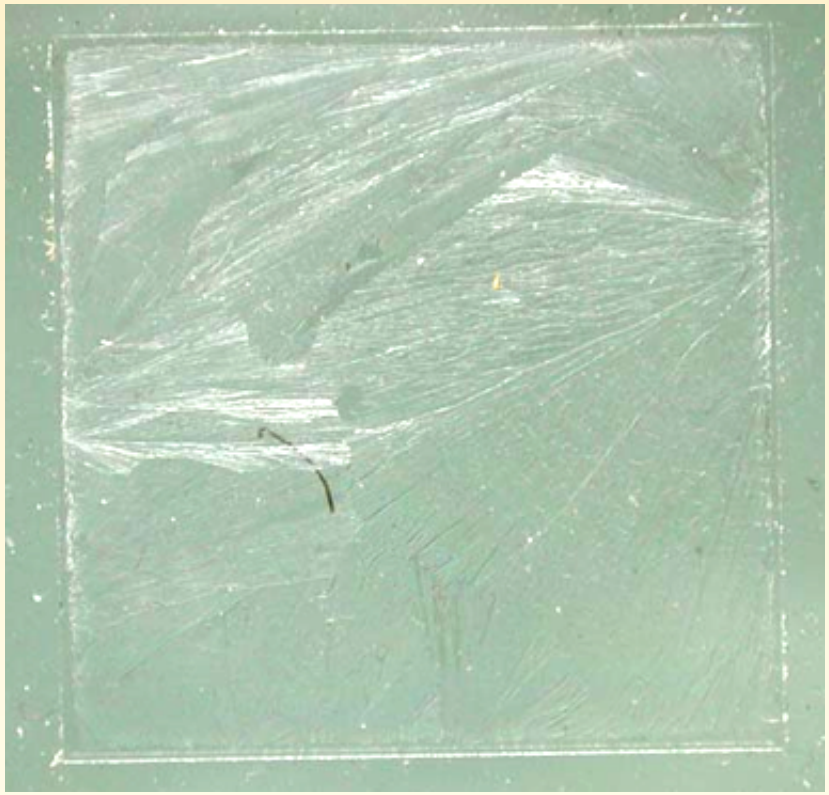
Overlapping pieces of Sellotape



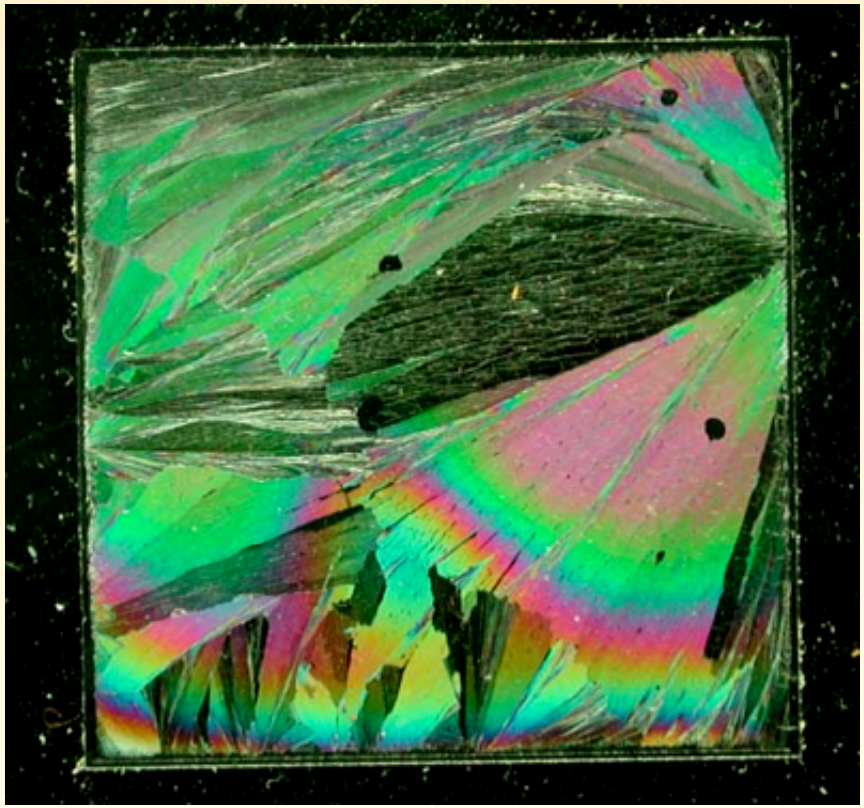
Crossed polars

4. Polarised Light - Example

Crystals of Acetanilide

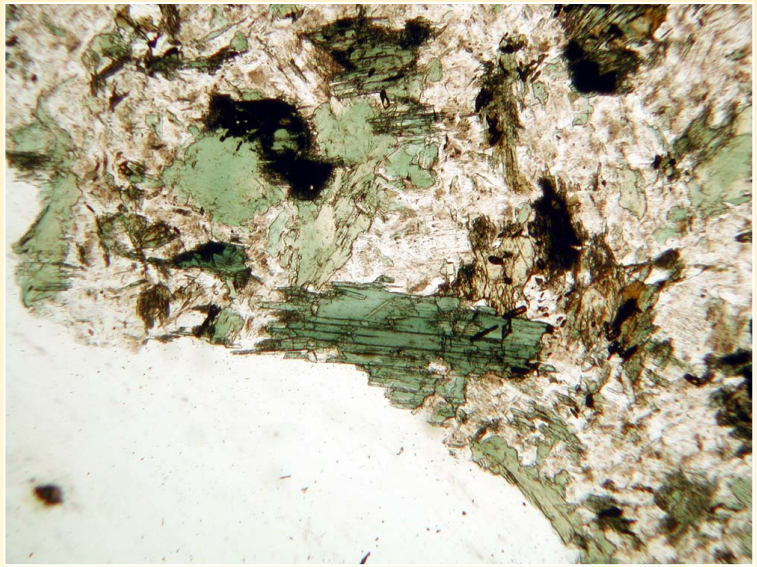


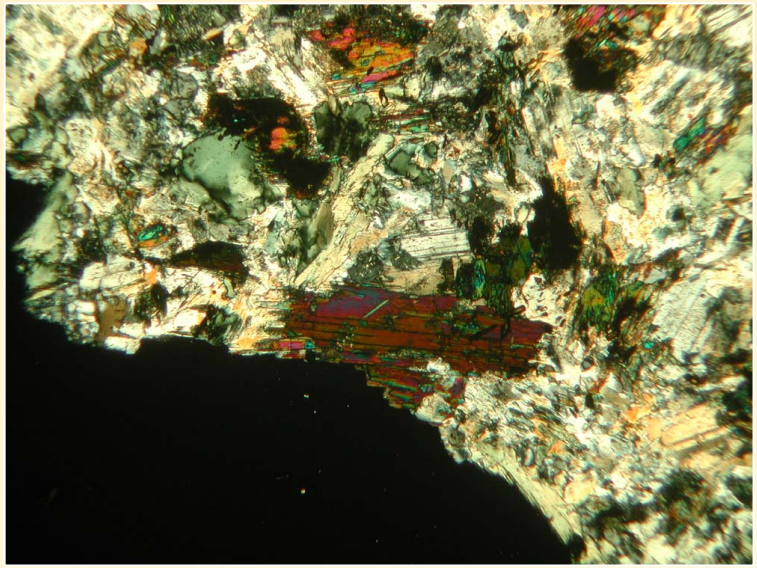
Natural light

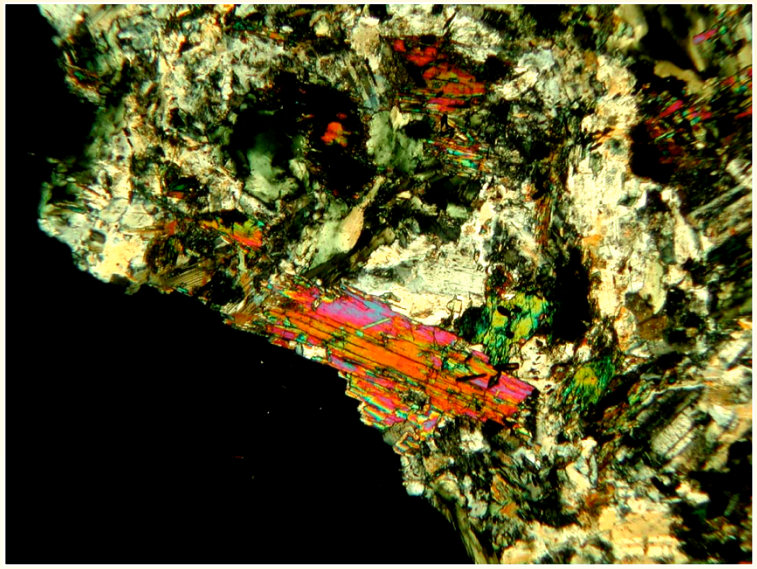


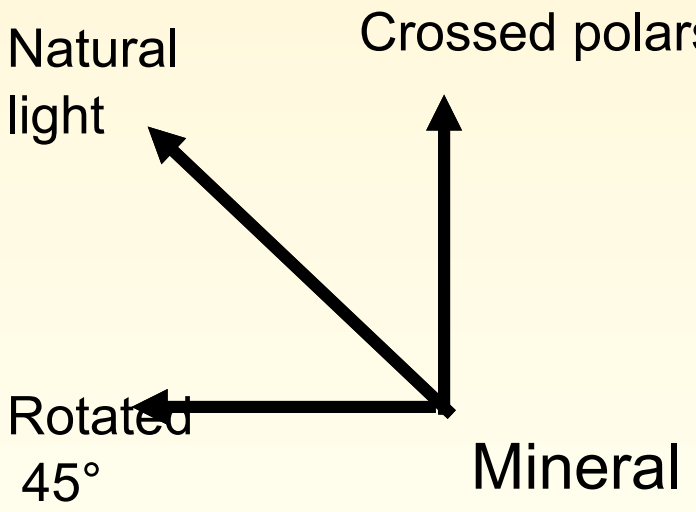
Crossed polars

4. Polarised Light - Example









Birefringence in Biological Materials

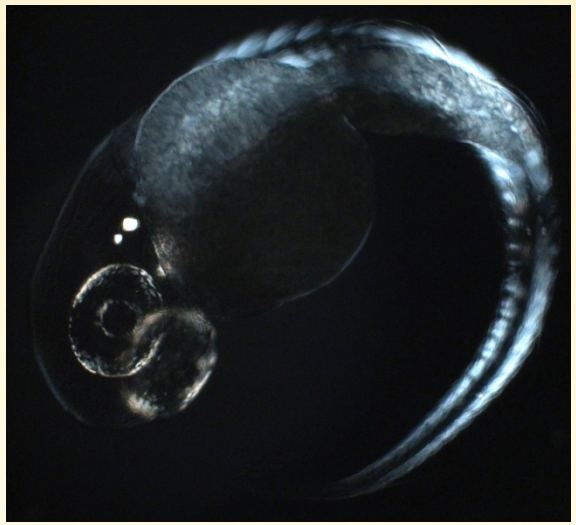
Anisotropic materials will generally be birefringent

What's anisotropic in the cell?

- Polymers: DNA, actin, microtubules
- Membranes
- Muscles:

BF

POL



Zebra fish embryo

Good for seeing **ordered structures in the cell**:

- Spindles, Other cytoskeletal structures, Membranes, Collagen

No staining required!

4. Polarized Light Microscopy

Try yourself

Setup:

- Köhler the microscope
- adjust polarizer and analyzer – crossed polars (black background)

Specimen:

- Hair
- Zebrafish (muscles)
- Honey (sugar crystals), starch, urea
- plasic material (tesa tape, plastic ruler)

Additional information:

- no plastic dishes, as it is birefringent. Only use glass, or glass bottom dishes (no lid).
- some objectives are especially suited for polarisation. They are labeled with “DIC” or labels are in red letters. All other objectives still work.

Wave Optics - Polarisation

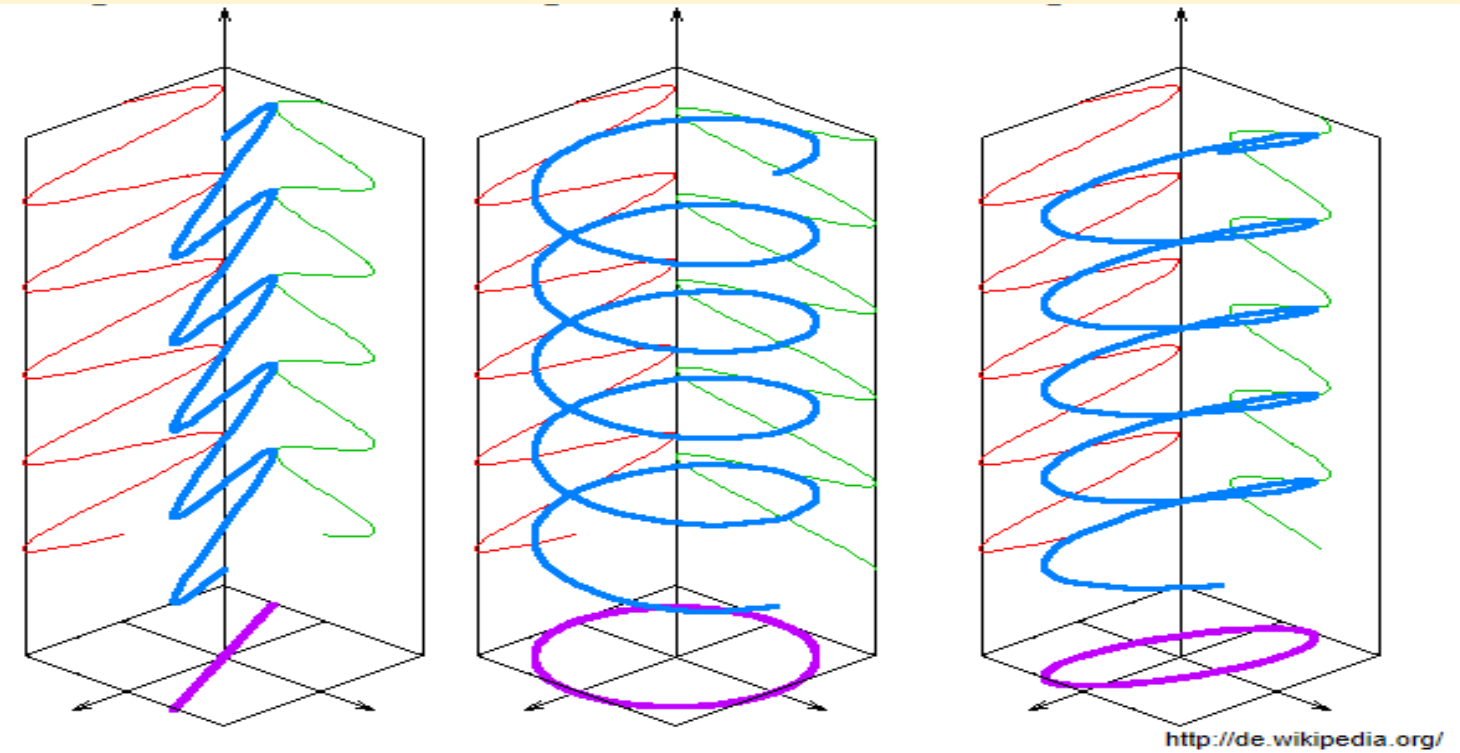
Polarized light as before is called **Linearly Polarized Light.**

... but there is more ...

superposition “in phase”
→ linearly polarized

superposition λ/4 “off phase”
→ circularly polarized

superposition α “off phase”
→ elliptically polarized



Modified from: http://www.univie.ac.at/mikroskopie/1_grundlagen/optik/wellenoptik/6_polarisation.htm