

Biophysics I

7. Luminescence

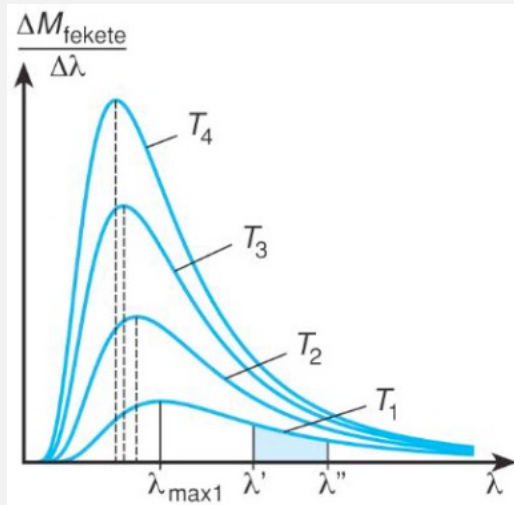
Liliom, Károly

18. 10. 2024.

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karoly.liliom.mta@gmail.com

Generation of light

Thermal radiation



Luminescence

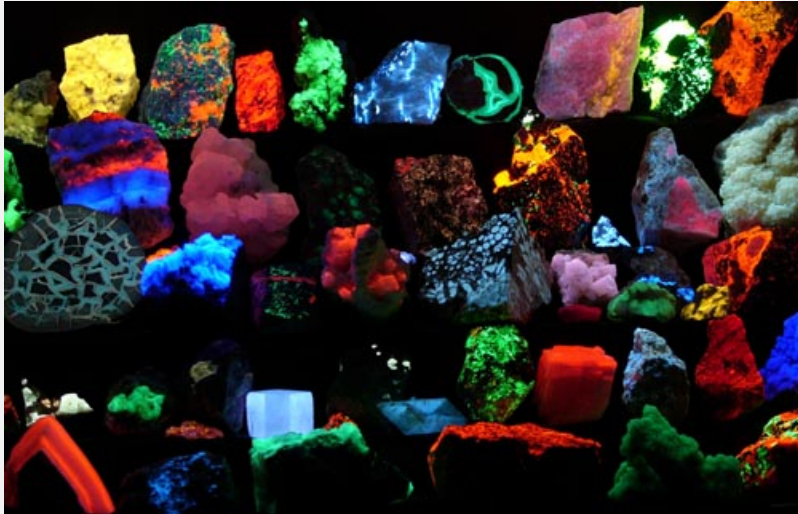


Laser

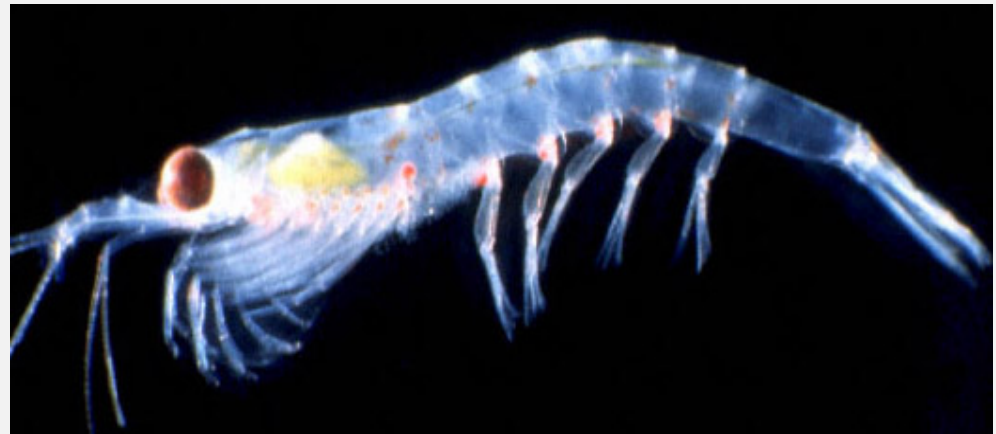


Luminescence: radiation released above the thermal radiation of a body (cold light).

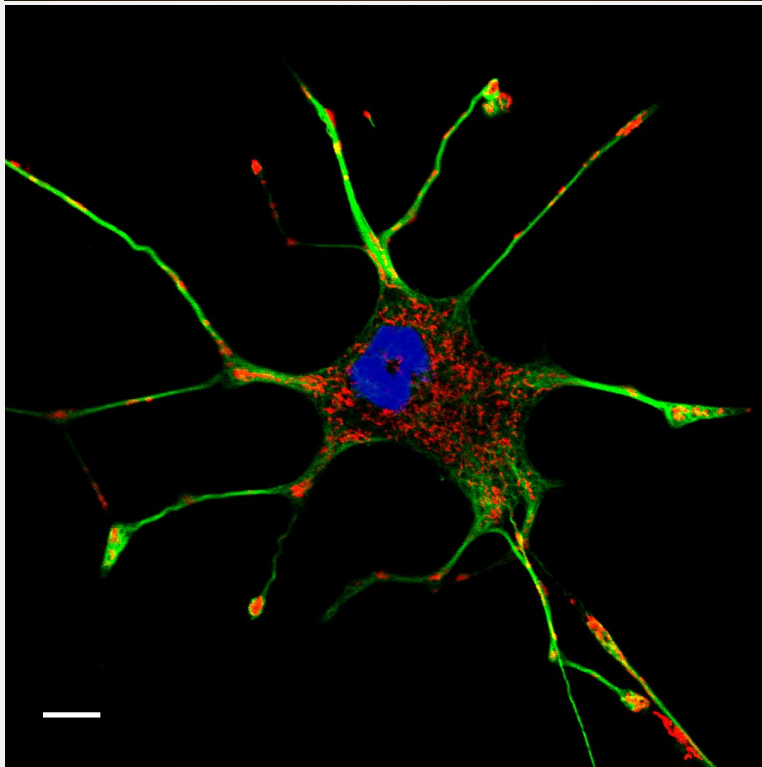
Luminescence in Nature



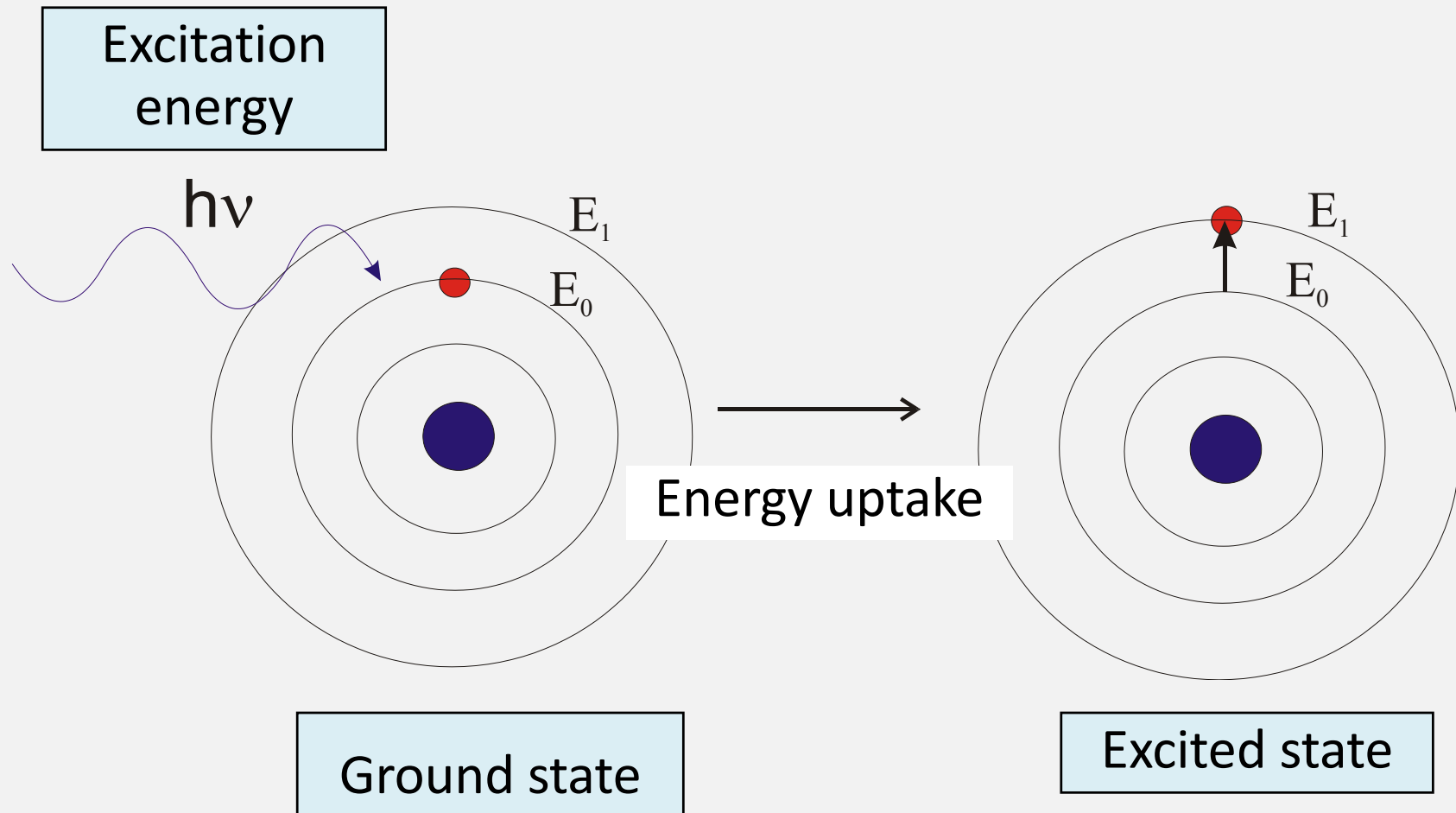
minerals, phyto- and zooplankton, jellyfish, plants...



Applied Luminescence

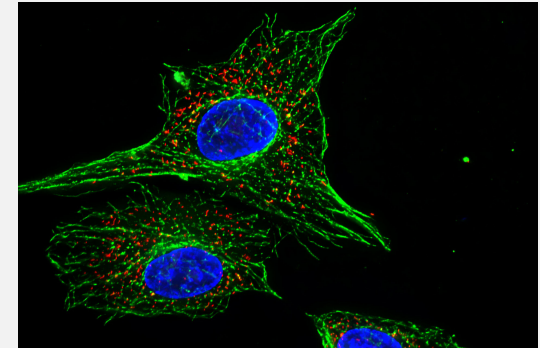


Let's consider a single atom



Typical excitation modes

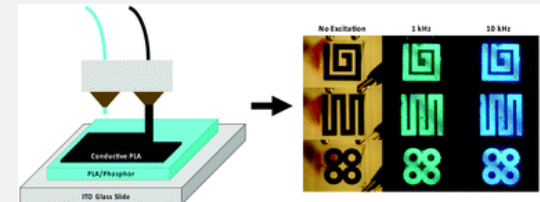
- absorption of photons: *photoluminescence*



- energy of a chemical reaction: *chemo/bioluminescence*



- high electric field or current: *electroluminescence*



- mechanical deformation: *triboluminescence*

mint candy

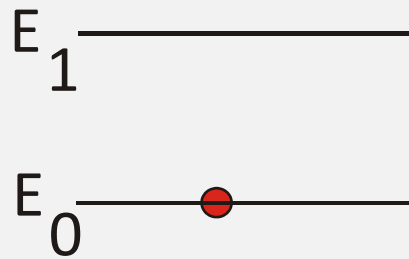


- heat: *thermoluminescence*

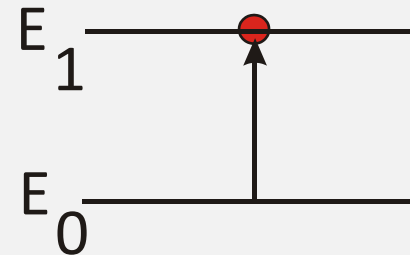
Wulfenit



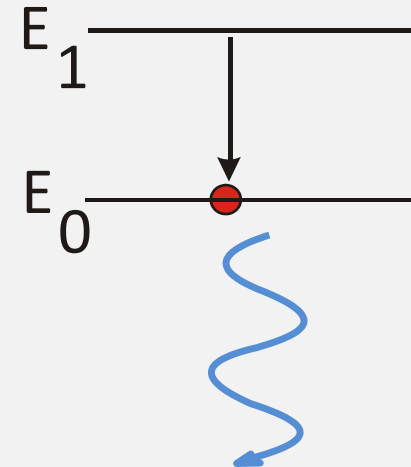
Mechanism of relaxation



excitation of an
external electron



spontaneous
relaxation of the
electron back to
ground state



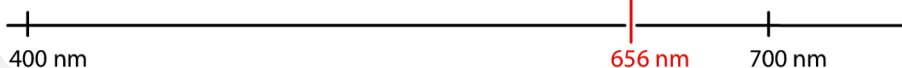
light emission

$$hf = E_1 - E_0$$

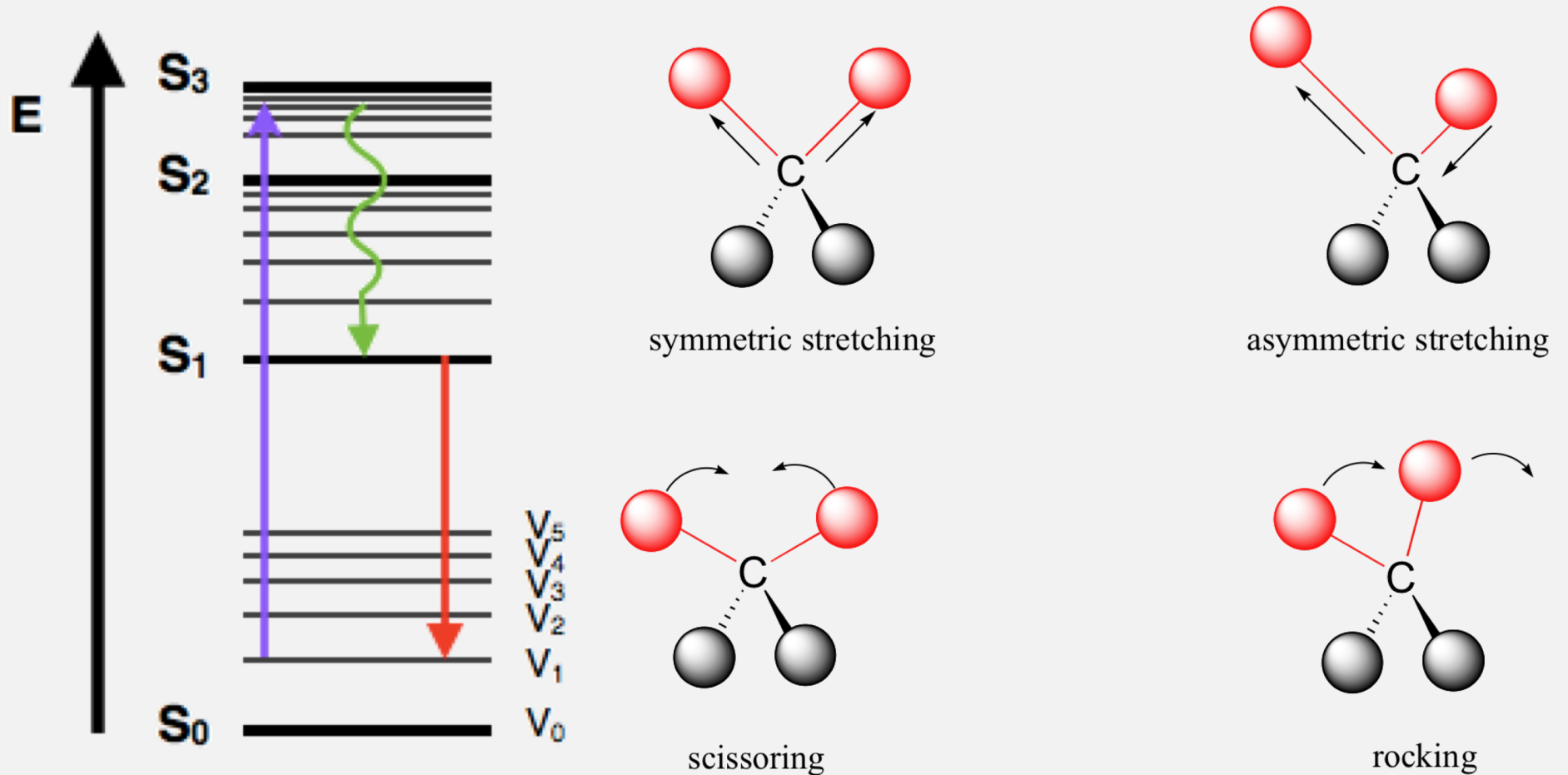
Absorption spectrum of Hydrogen



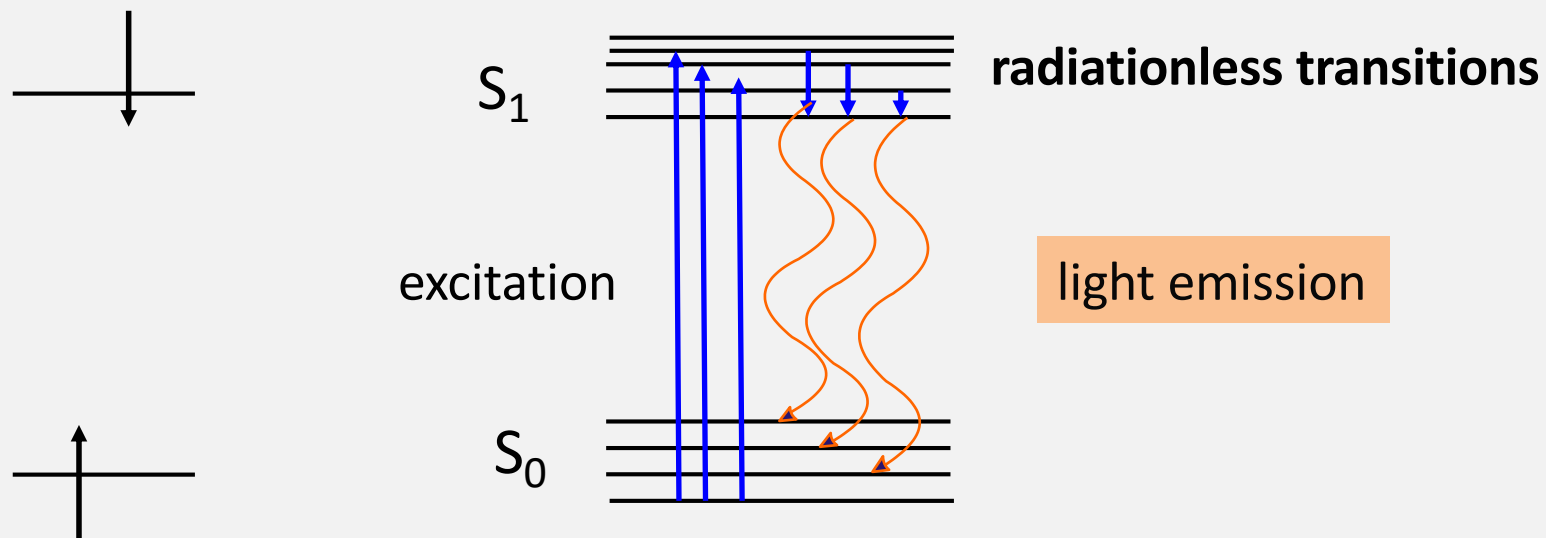
Emission spectrum of Hydrogen



Energy levels in Molecules



Mechanism of Fluorescence

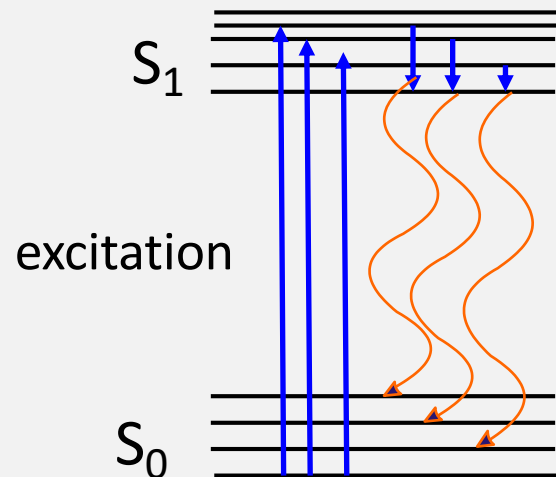


Singlet state:

paired electrons
with net spin of 0
(multiplicity of 1)

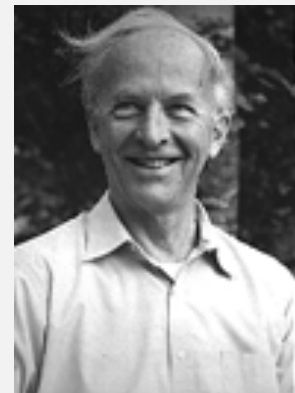
Fluorescence

**Excitation and relaxation (light emission) in singlet states
– no change in spin state!**



Kasha's rule:

light emission emanates from the relaxation of the lowest vibrational level of the first excited state to ground state



$$E_{\text{excitation}} \geq E_{\text{fluorescence}}$$

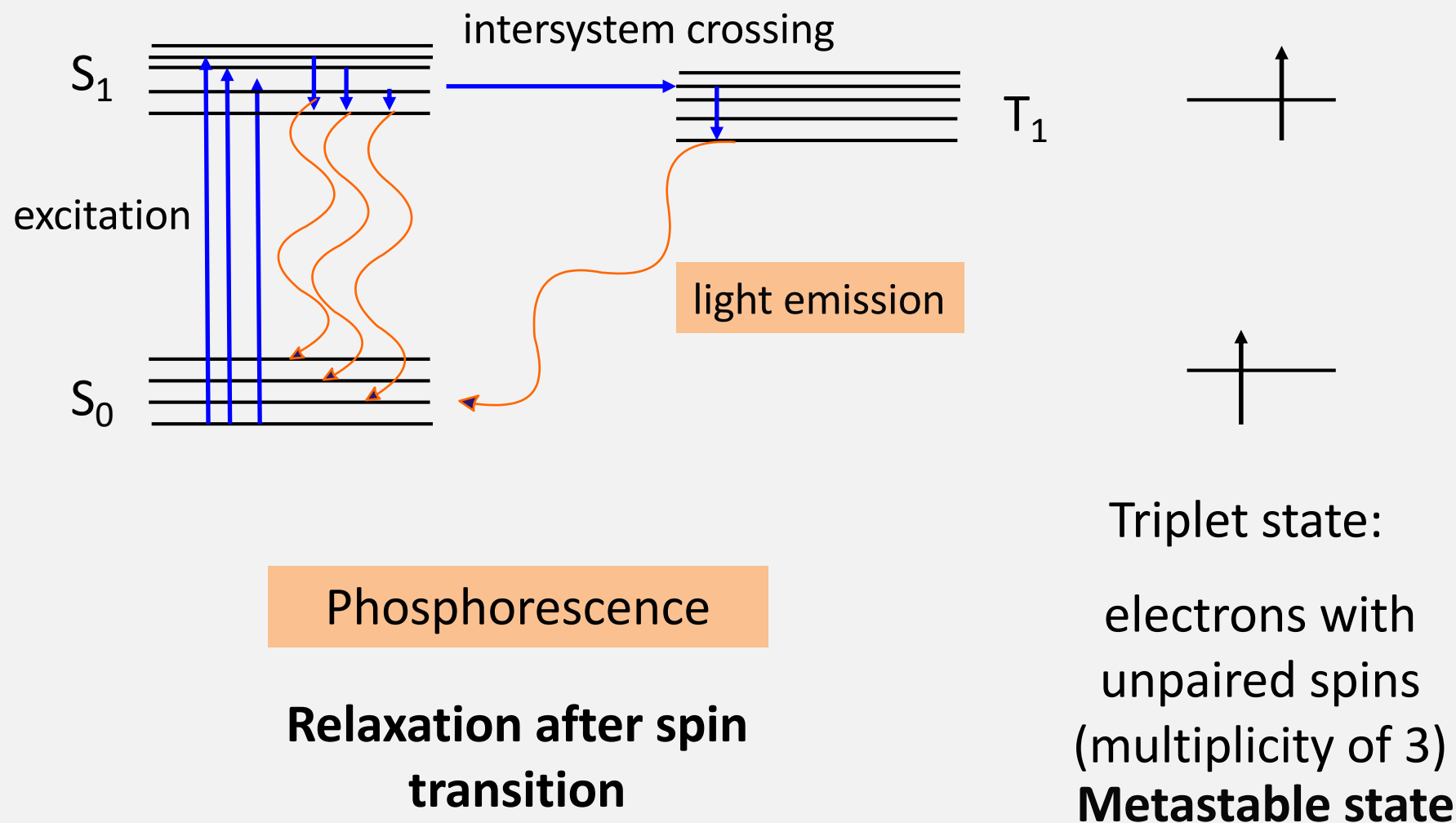
$$\lambda_{\text{excitation}} \leq \lambda_{\text{fluorescence}}$$

Stokes's shift

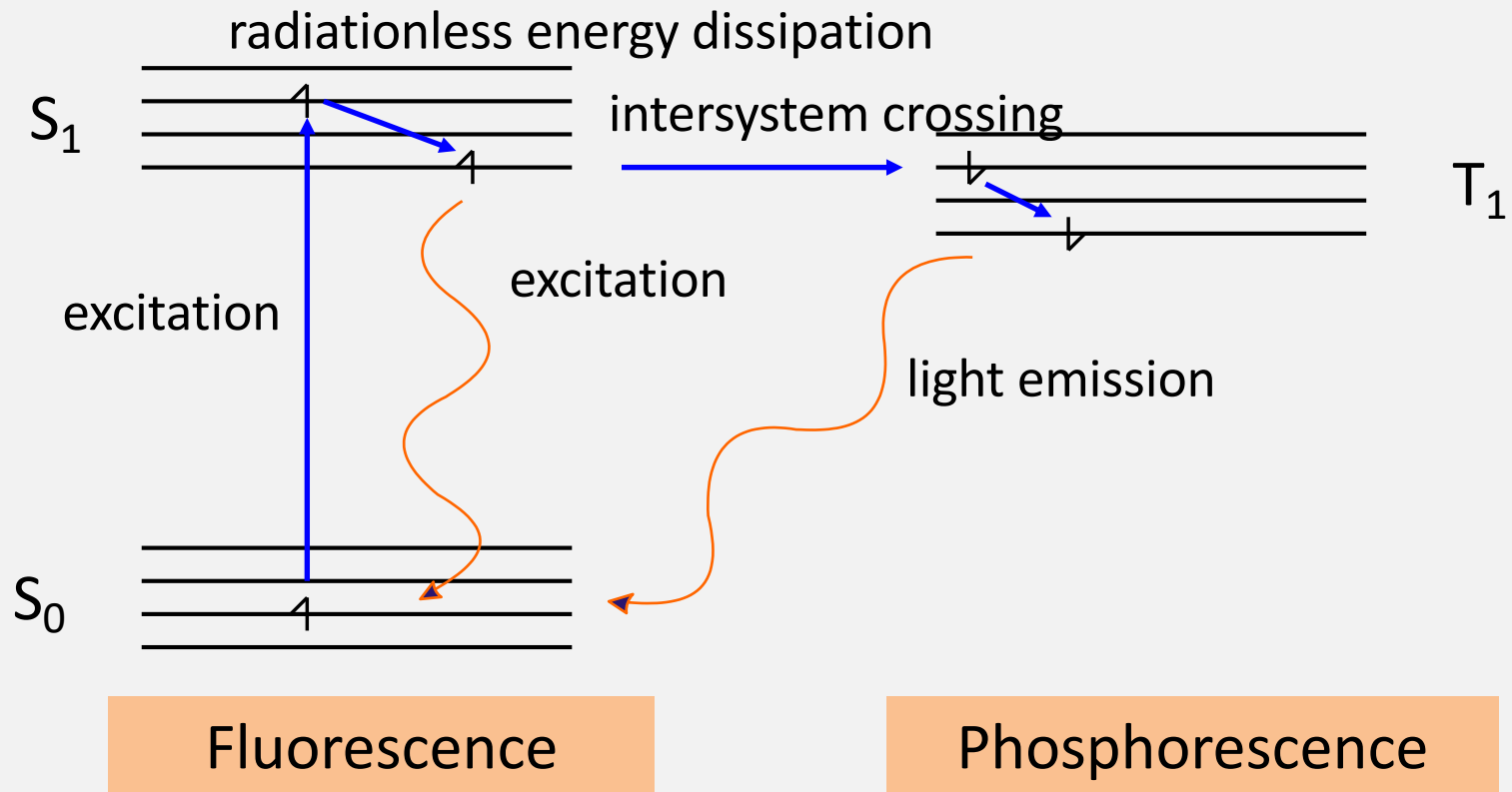
$$E = h^*c/\lambda$$



Mechanism of Phosphorescence



Energy-relationship of luminescence modes



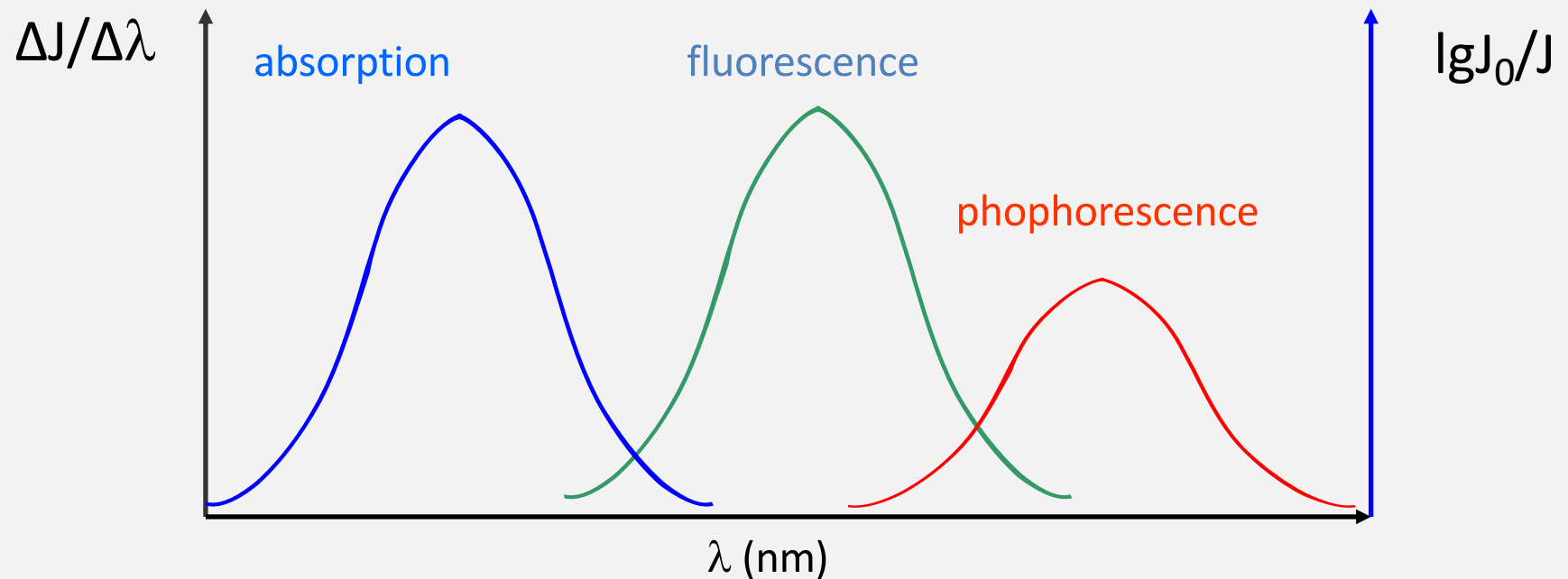
Stokes's shift

$$E_{\text{excitation}} \geq E_{\text{fluorescence}} > E_{\text{phosphorescence}}$$

$$\lambda_{\text{excitation}} \leq \lambda_{\text{fluorescence}} < \lambda_{\text{phosphorescence}}$$

Characteristics of emitted light

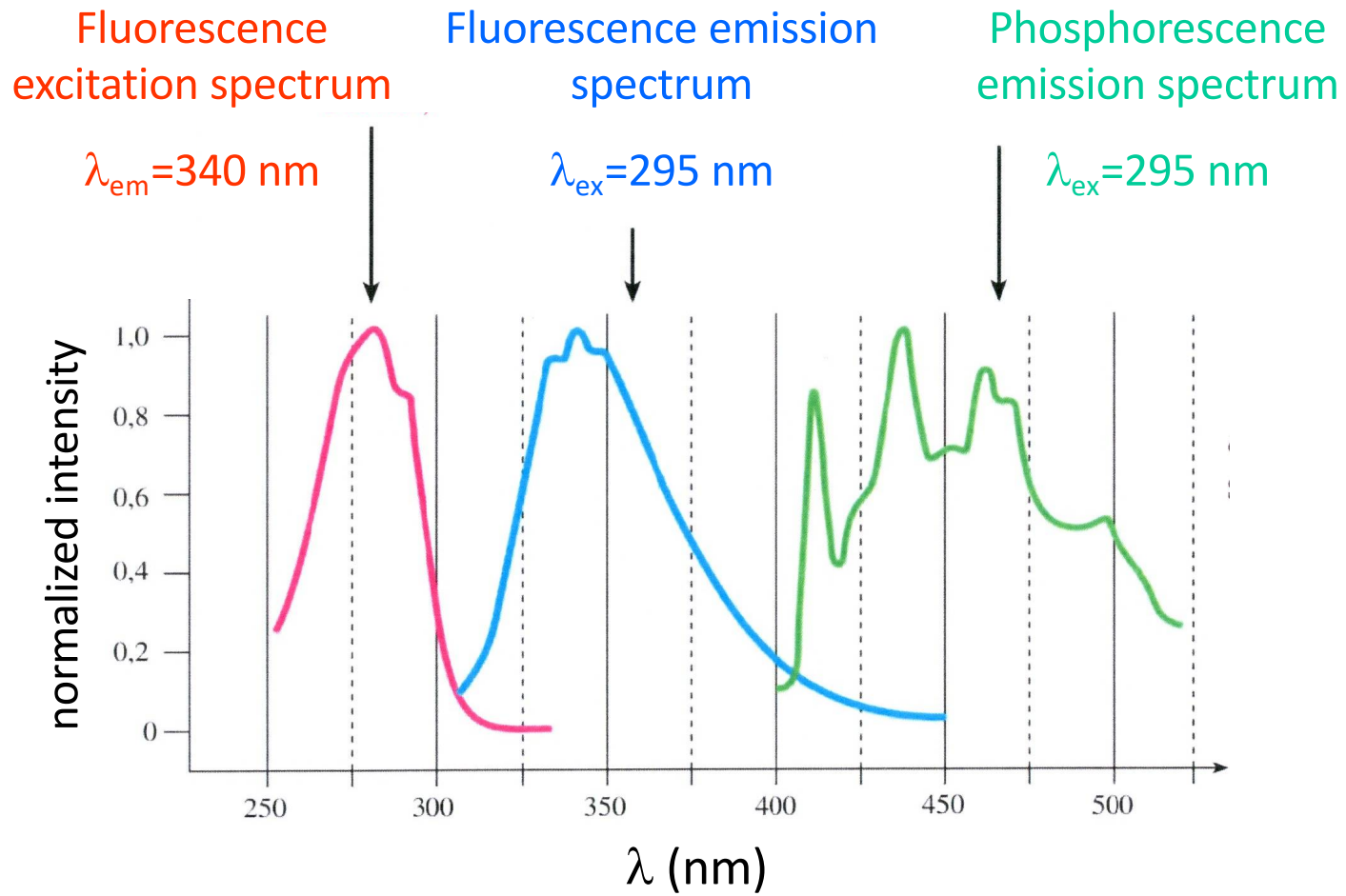
Band-spectrum in case of molecules



$$\lambda_{\text{excitation}} \leq \lambda_{\text{fluorescence}} < \lambda_{\text{phosphorescence}}$$

Stokes's shift

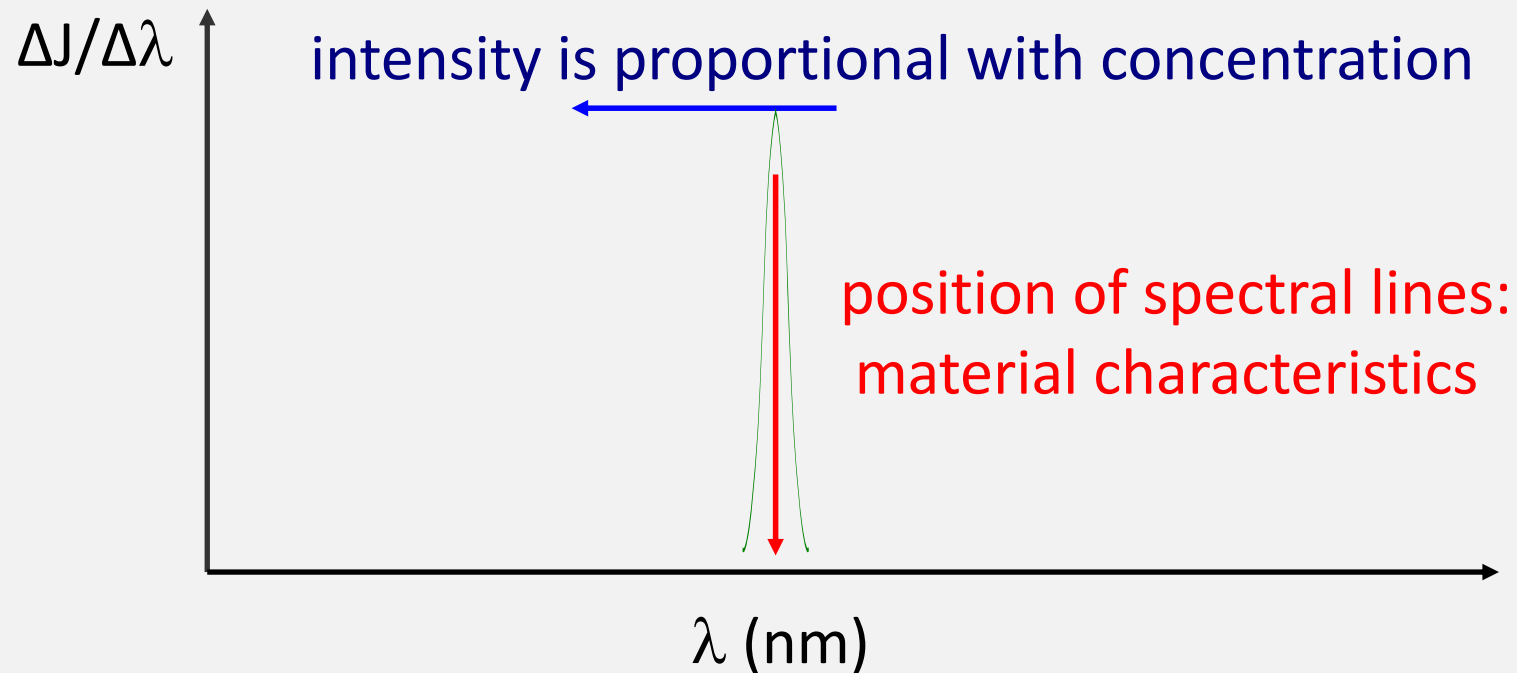
Spectra of tryptophan



Characteristics of emitted light

Spectrum – distribution of intensities along wavelength

Line-spectrum in the case of atoms:

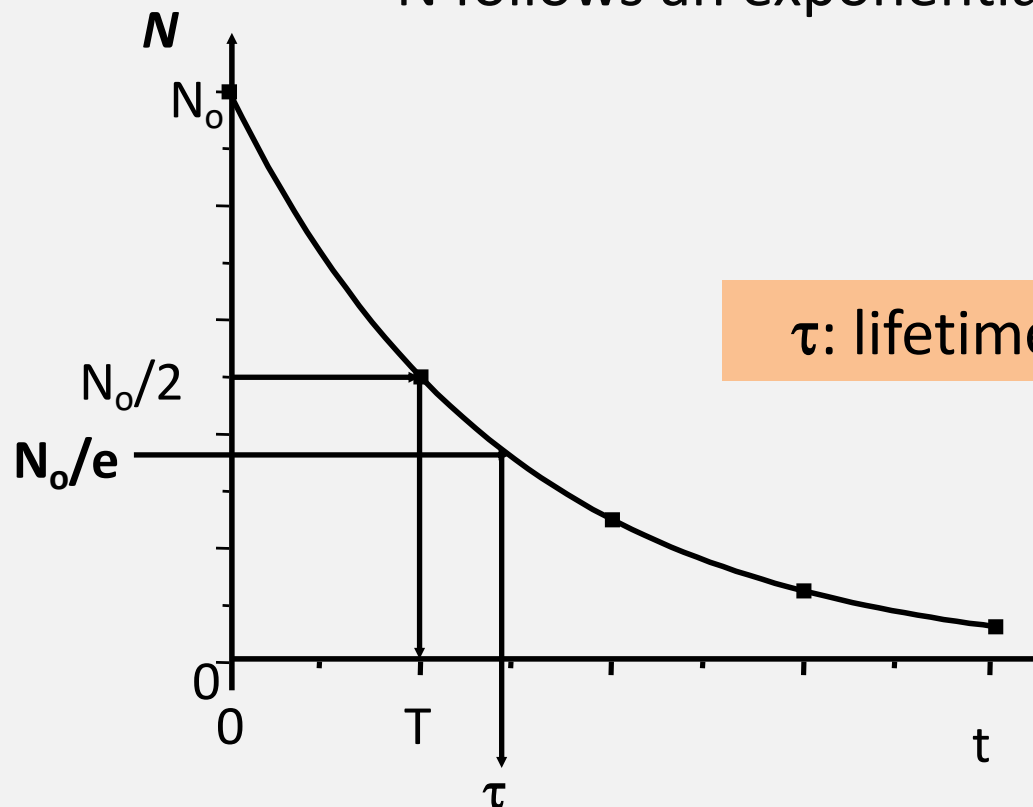


Excited-state lifetime

Number of excited electrons \longrightarrow $N = N_0 e^{-\frac{t}{\tau}}$

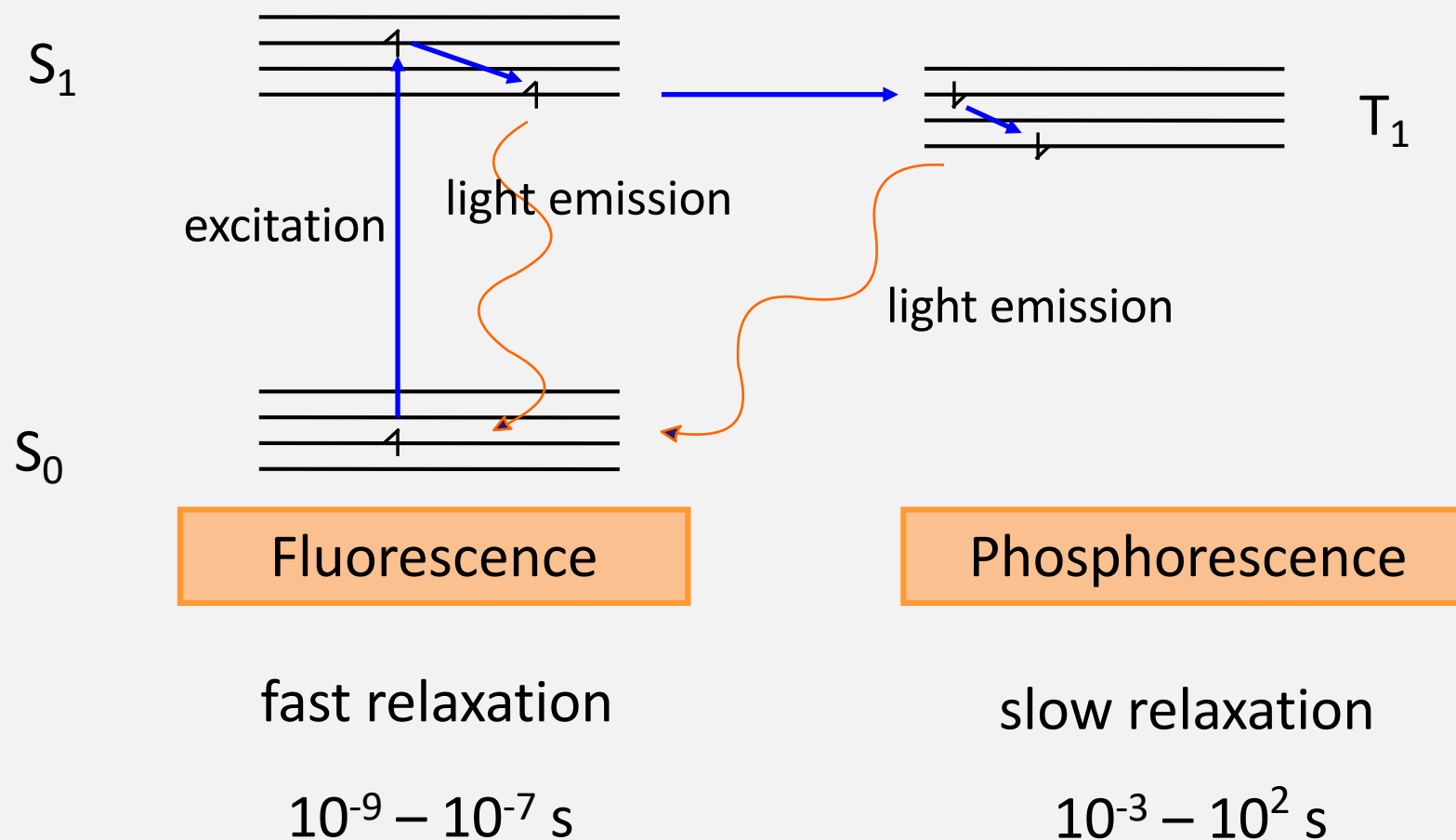
time after excitation \nearrow

N follows an exponential decay function.



τ : lifetime, T : half-life

Typical excited-state lifetimes



Is excitation always followed by photon emission?

- Excited state relaxation can be caused by mechanisms other than photon emission and are therefore often called “non-radiative transitions”.
- These can include: chemical reaction, dynamic collisional quenching, near-field dipole-dipole interactions, internal conversion and intersystem crossing.

Is excitation always followed by photon emission?

Fluorescence quantum yield (Q_F)

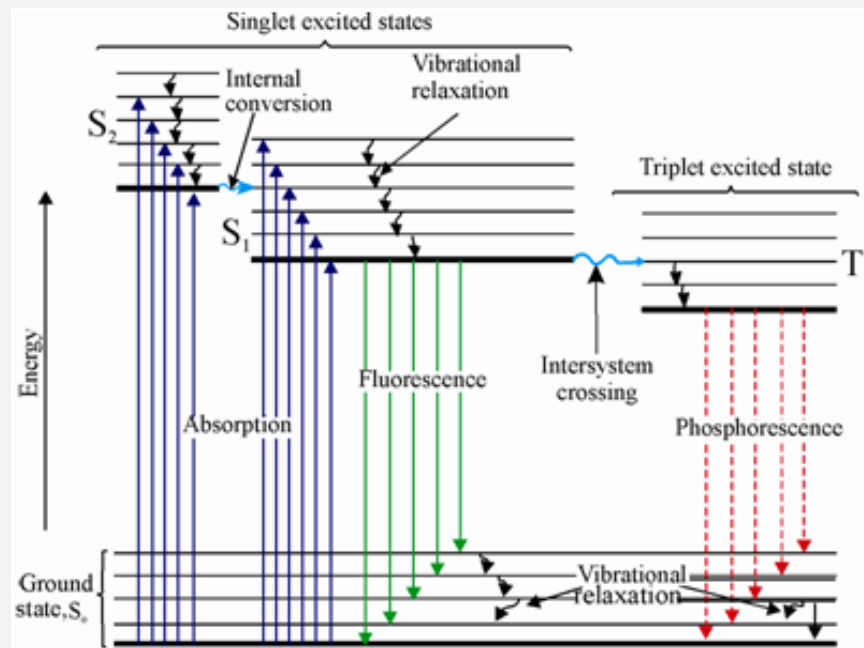
Q_F = number of emitted photons / number of absorbed photons

$$Q_F \leq 1$$

Luminescence summary

Types of luminescence:

- fluorescence
- phosphorescence



Characteristics:

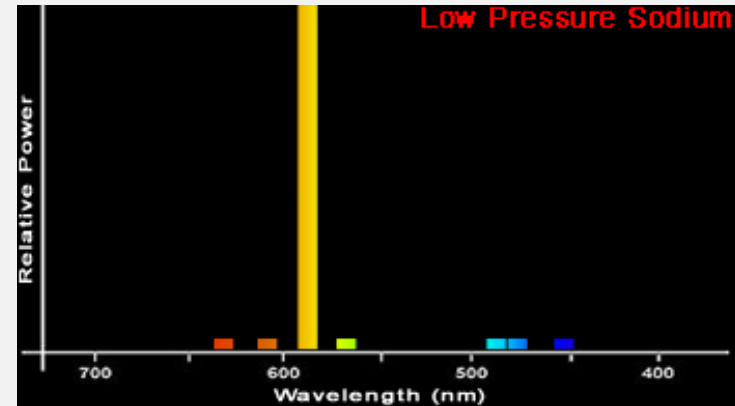
- emission spectrum:
 - type
 - maximum λ position
 - amplitude
- lifetime
- quantum yield

Application fields of luminescence

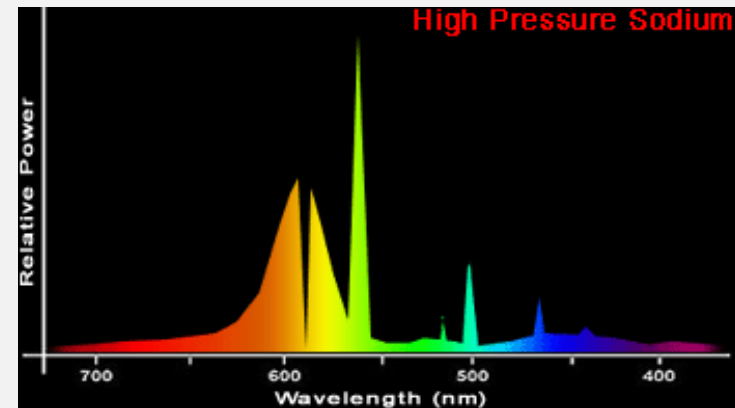
- light sources
- concentration determination
- fluorescence spectroscopy
- fluorescence microscopy
- dosimetry
- structure determination
- cell/tissue labeling
- safety control ... many more

Luminescent light sources

Metal-vapor lamps

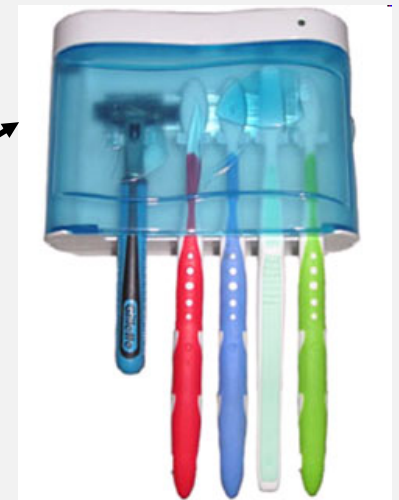
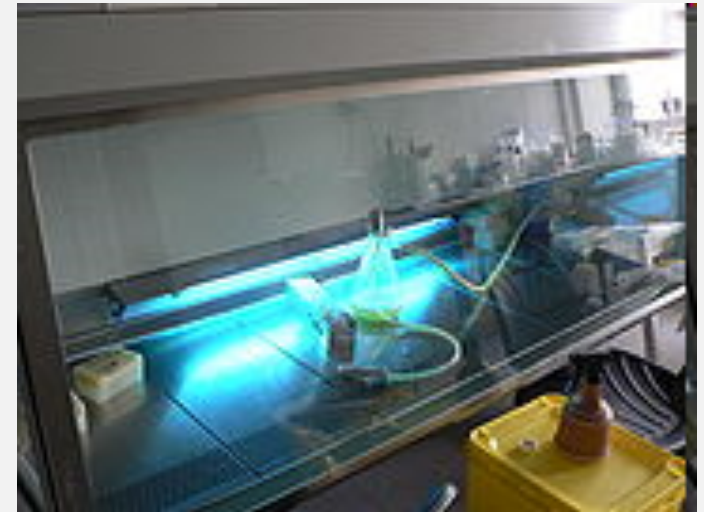
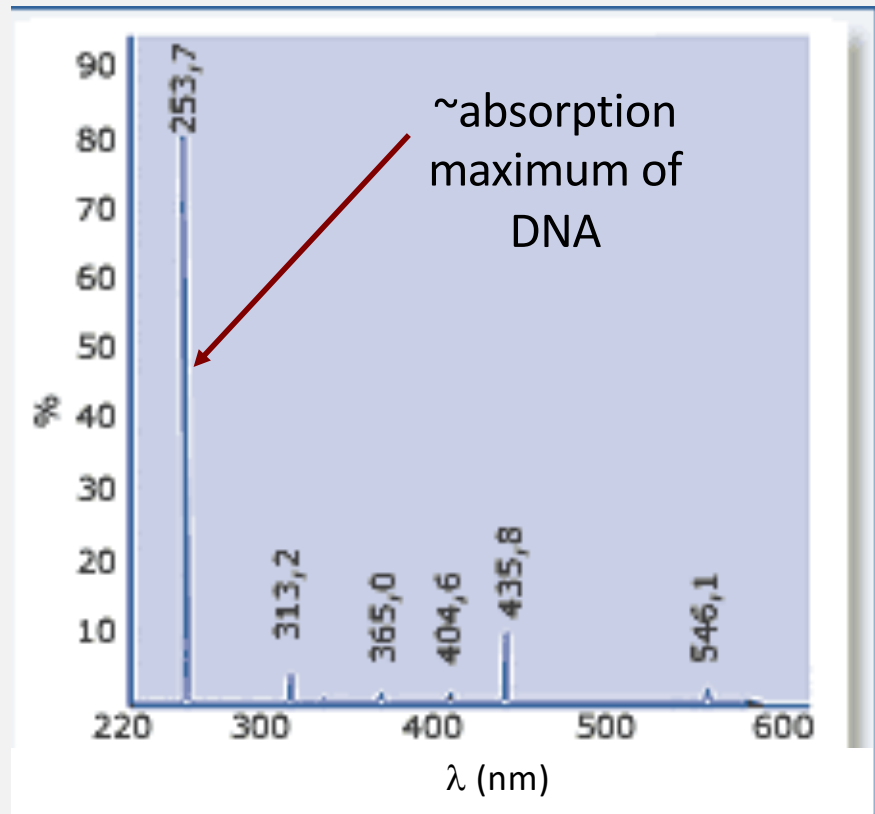


Low-pressure Na-vapor lamp



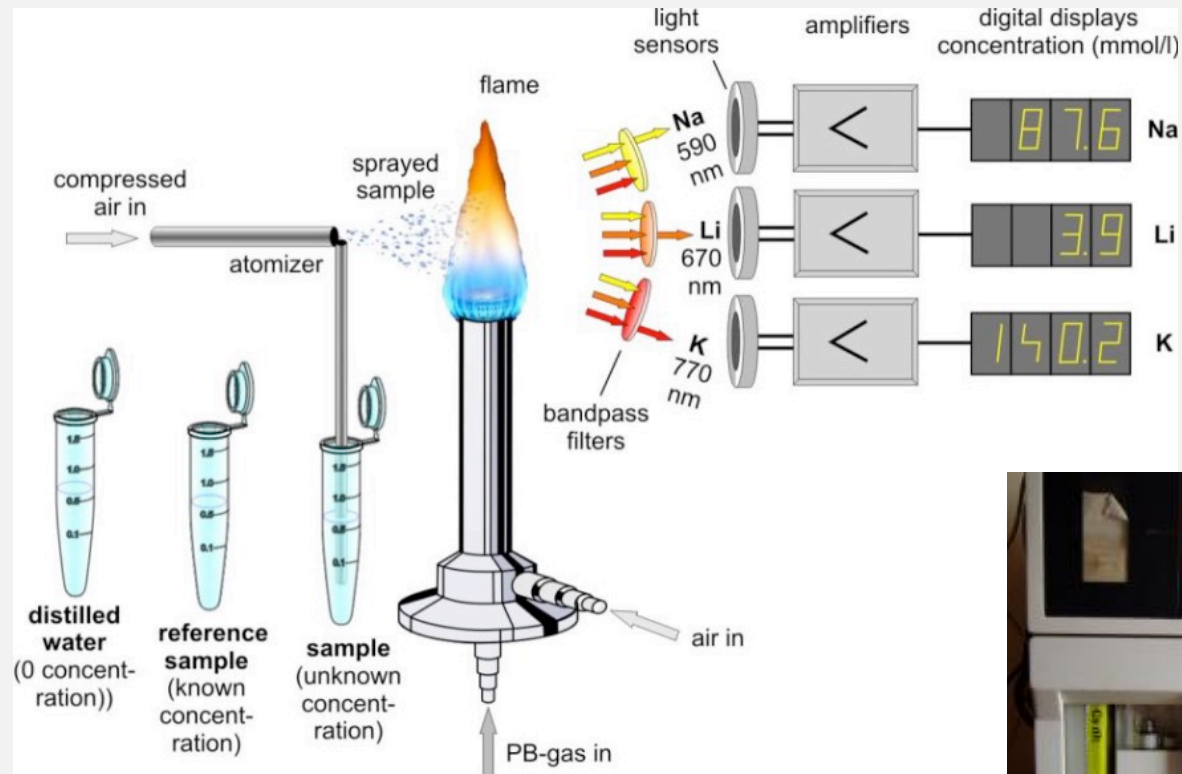
High-pressure Na-vapor lamp

Low-pressure Hg-vapor lamp



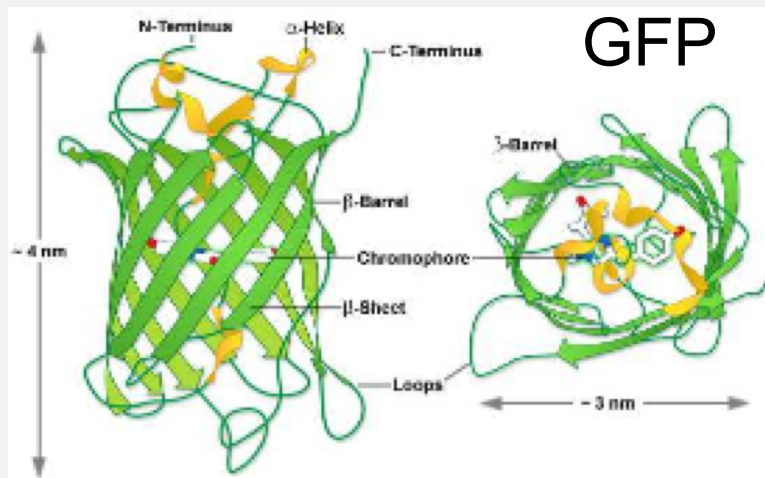
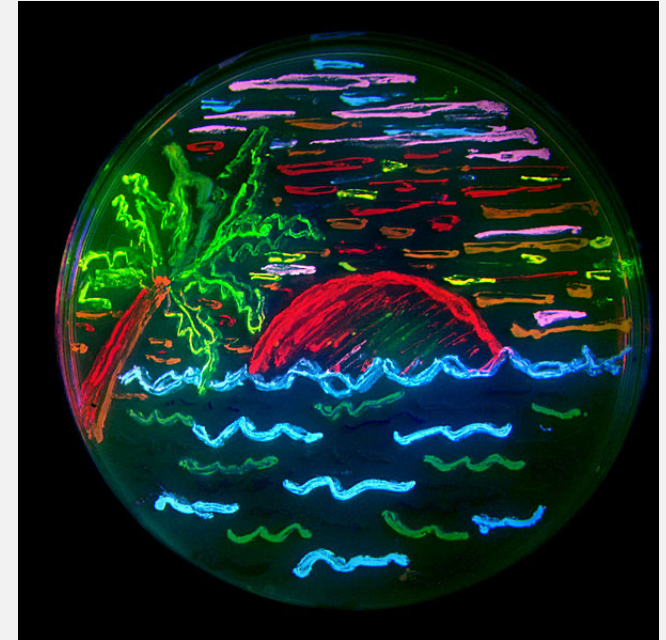
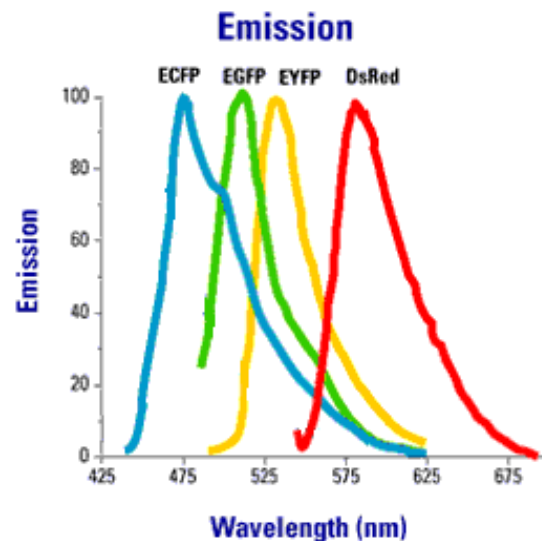
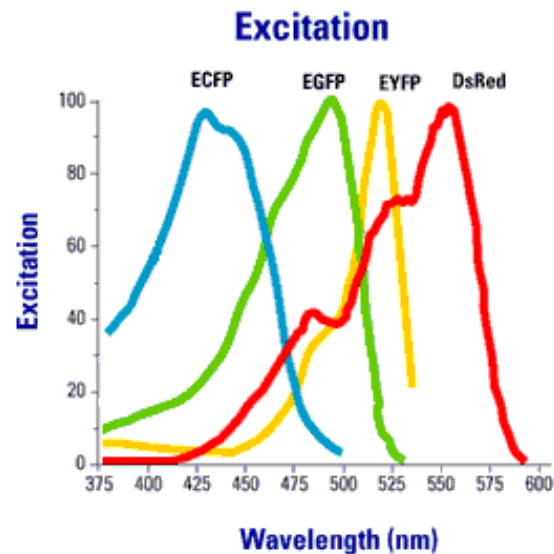
application: germicid lamp

Flame photometer

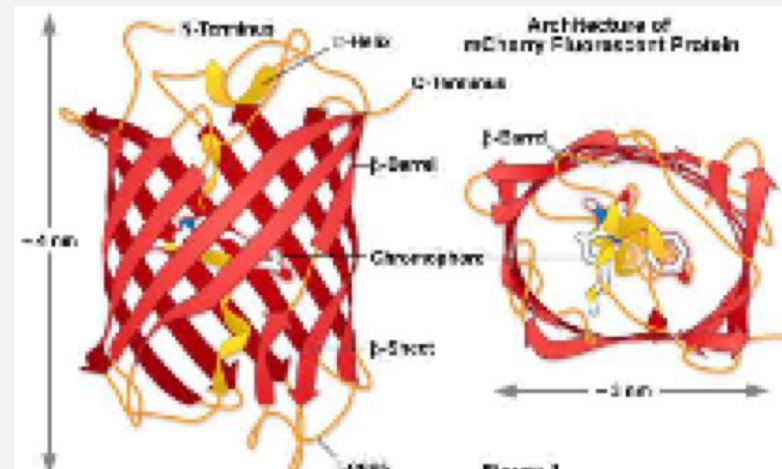


Quantitative determination of K^+ , Li^+ és Na^+ ions

Fluorescence-based methods are wide-spread in medical research and diagnostics

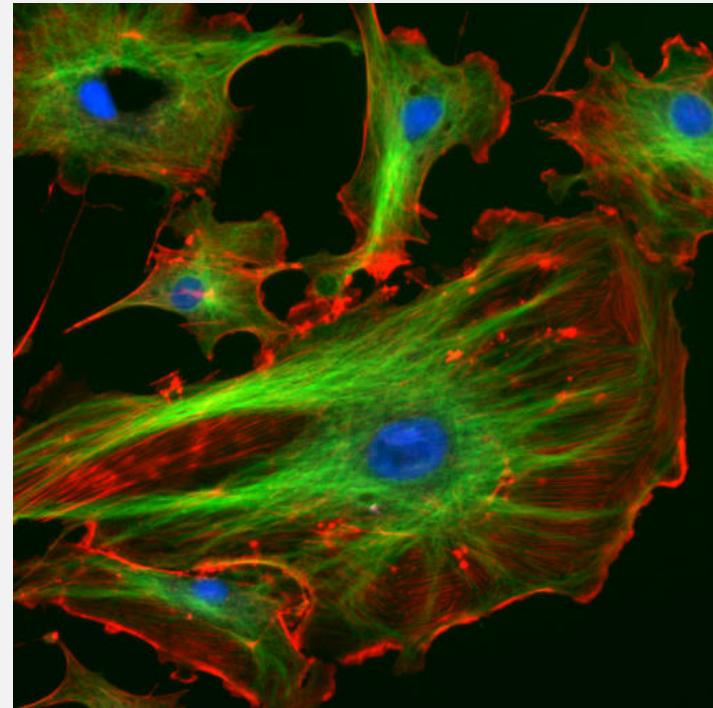
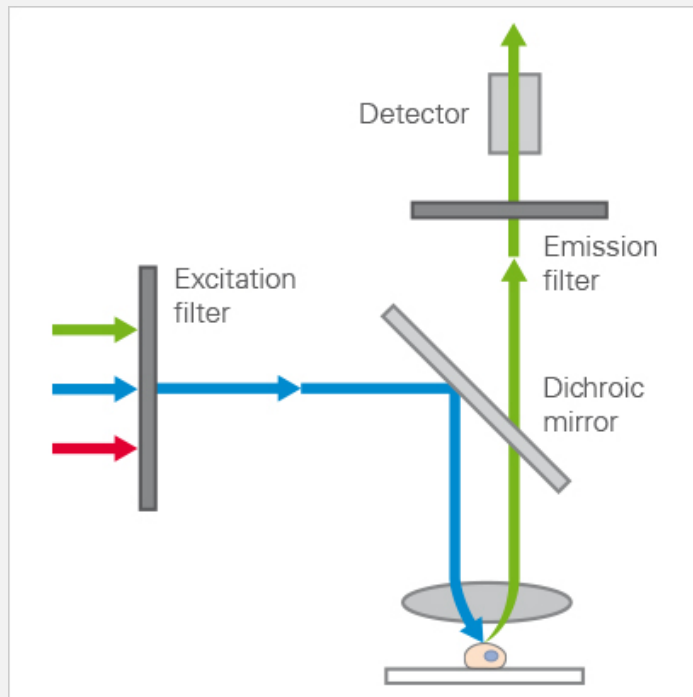


GFP (jellyfish)

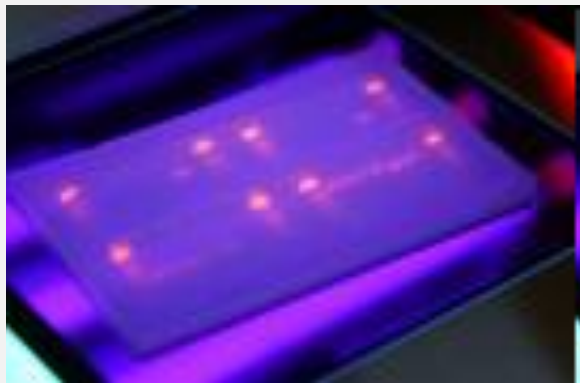


DsRed (red coral)

Fluorescence microscopy



Many applications in biomedical research



protein/DNA staining



fluorescent animal models

Fluorescent quantum dots (QD)

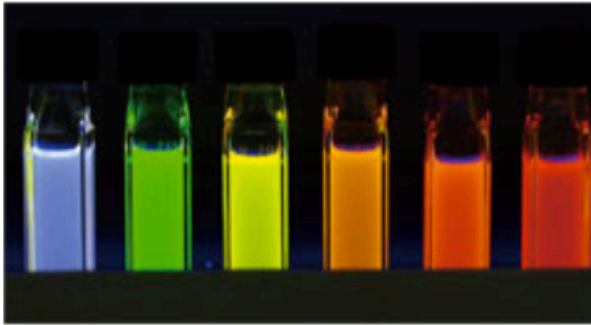
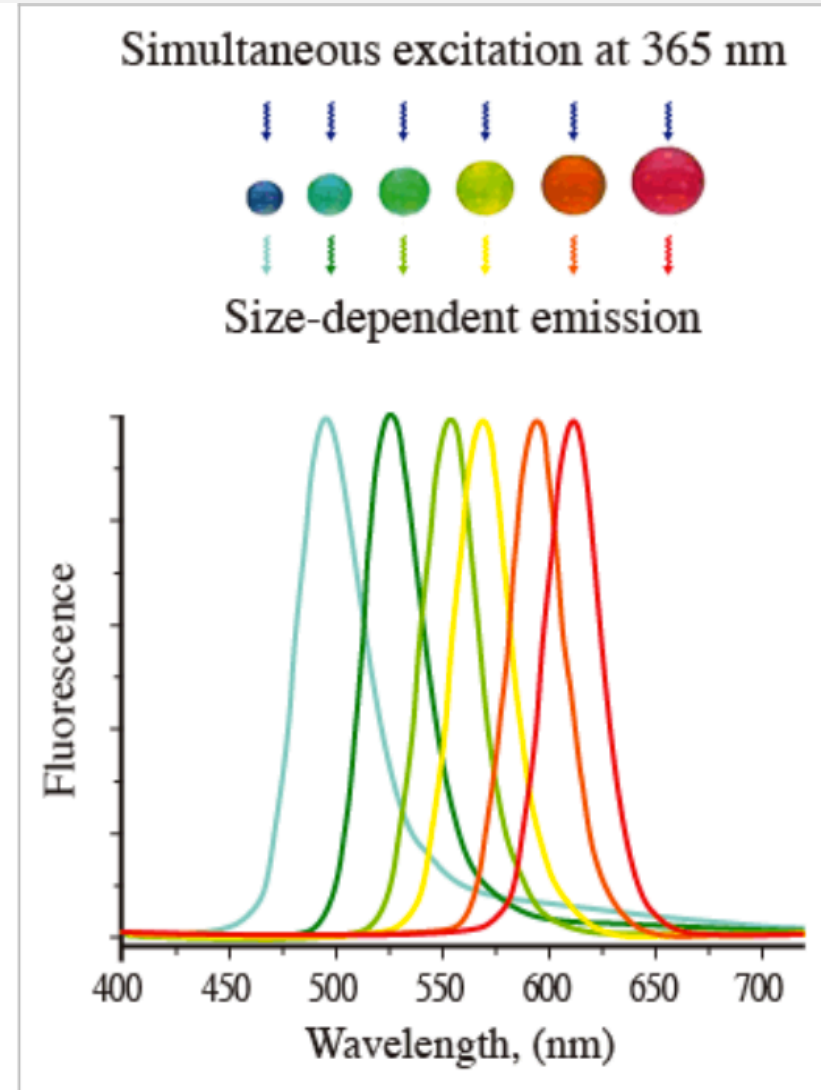
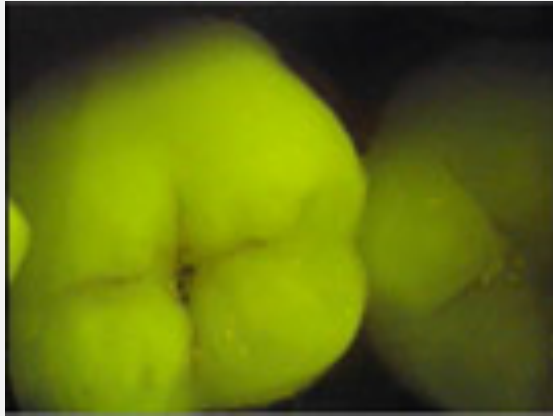


Figure 1: Fluorescence emitted from quantum dots. Blue fluorescence can be emitted from small particles of approximately 2 nm in diameter, green from ~3 nm particles, yellow from ~4 nm particles, and red from large particles of ~5 nm. The wavelength of the excitation light is 365 nm.

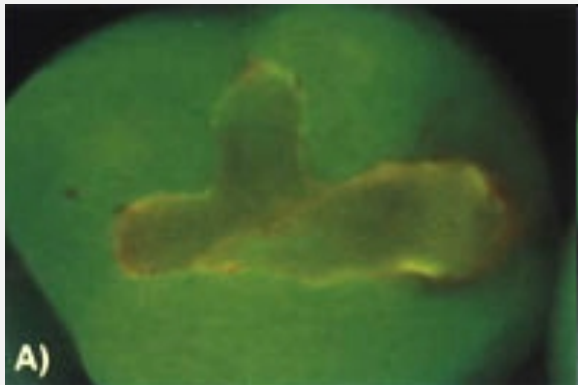
Excitation spectra overlap to ~400 nm, so only one excitation wavelength is proper to a set of QDs.



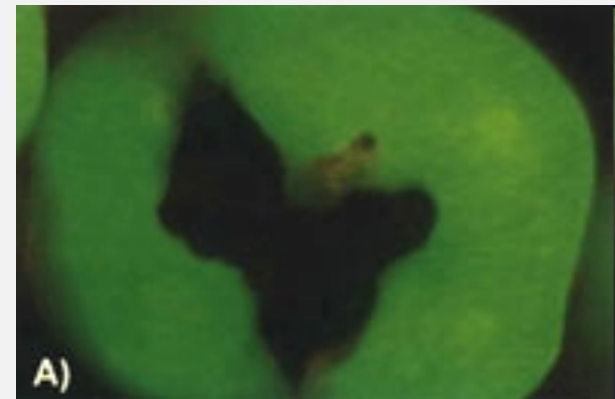
Applications in dental medicine



Auto-fluorescence of teeth. When teeth are illuminated with high intensity blue light they will start to emit light in green.



Red fluorescence indicates the activity of cariogenic bacteria



amalgam restoration

0 – 14	No special measures.
15 – 20	Usual prophylactic measures.
21 – 30	More intensive prophylaxis or restoration: indication is dependent on: *Caries activity. *Caries risk. * Recall interval, etc.
from 30	Restoration and more intensive prophylaxis.

KaVo DIAGNOdent
- How it Functions

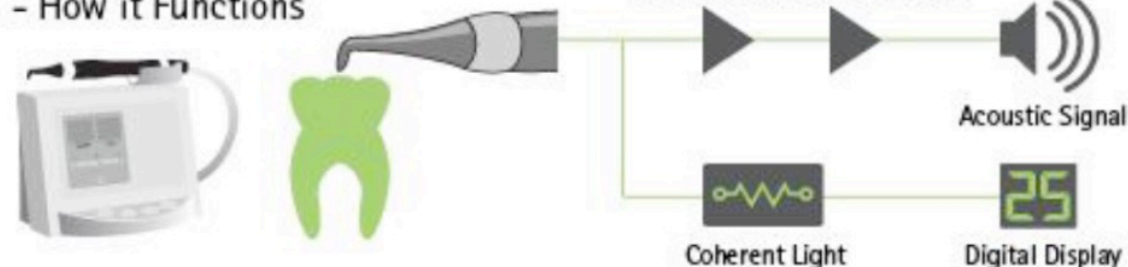


Figure (5) Spectra camera with spacer on (Kurtzman, 2010).

Table 2: Interpretation of Spectra data (Kurtzman, 2010).

Displayed Color	GREEN → BLUE → RED → ORANGE → YELLOW				
Displayed Number	1 —————→ 5				
Depth of Involvement	Sound Enamel	Initial Enamel Caries	Deep Enamel Caries	Initial Dentin Caries	Deep Dentin Caries



SOPROCARE. (A) Carious lesion invisible in DAYLIGHT mode. (B) Carious lesion visible in CARIO mode

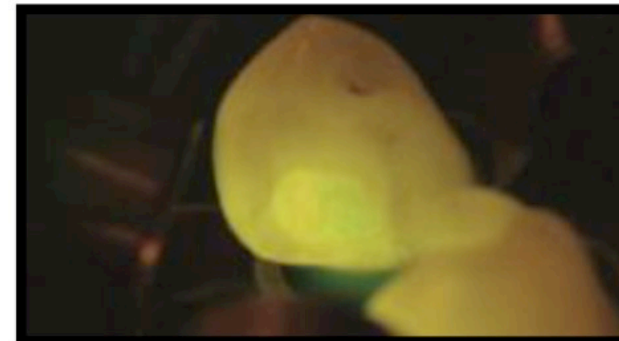
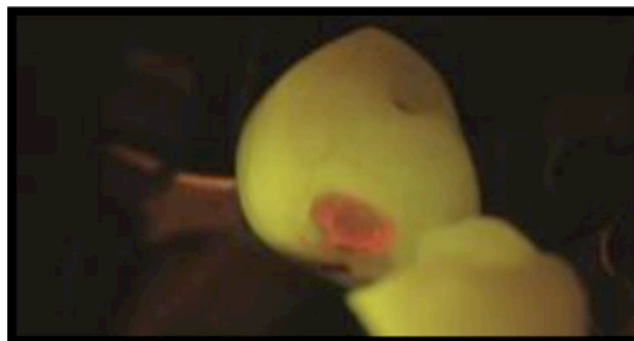
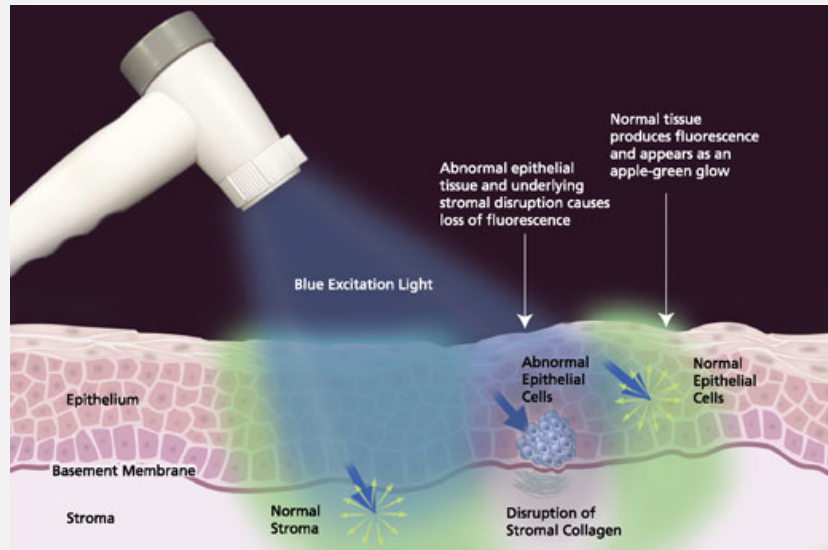
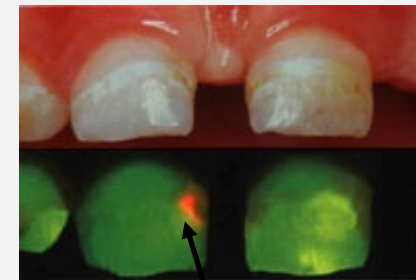


Figure (8) Photos showed cavity illumination with Facelight before and after caries excavation (21).

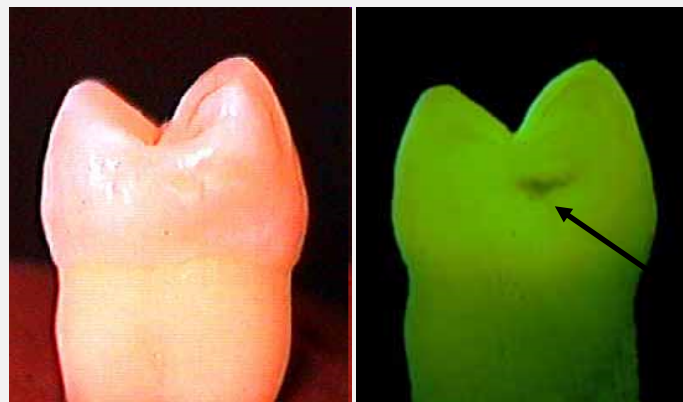


Healthy and malignant tissues -
different fluorescent properties

native and fluorescent
images



Active caries



native and fluorescent
images

caries

Checklist:

Luminescence

ground- and excited states

modes of excitation

Jablonski diagram

Fluorescence

Phosphorescence

Kasha's rule

Stokes's shift

lifetime

quantum yield

applications

Damjanovich, Fidy, Szöllősi: Medical Biophysics

II. 2.2

2.2.4

2.2.6

VI.3.3

3.3.1

3.3.2

3.3.3