

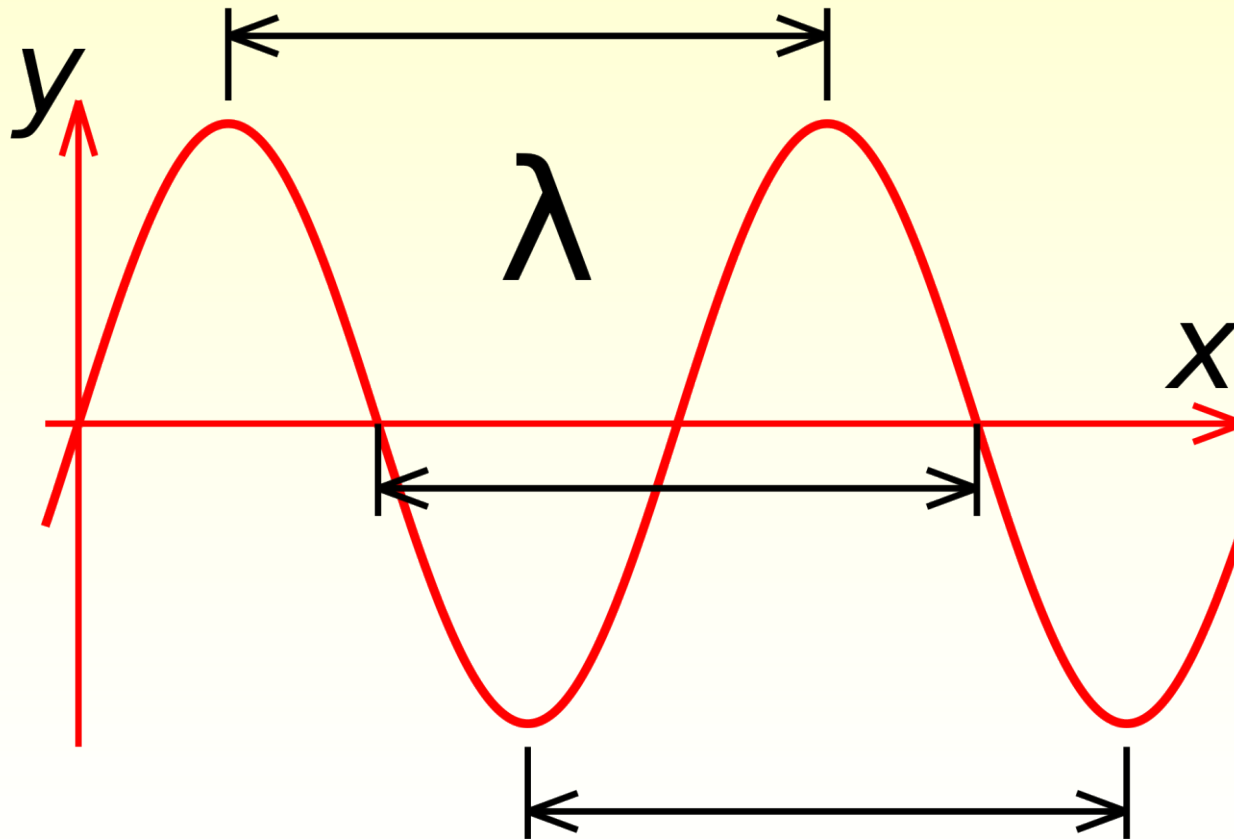
Spectroscopic methods of analysis

Spectroscopy

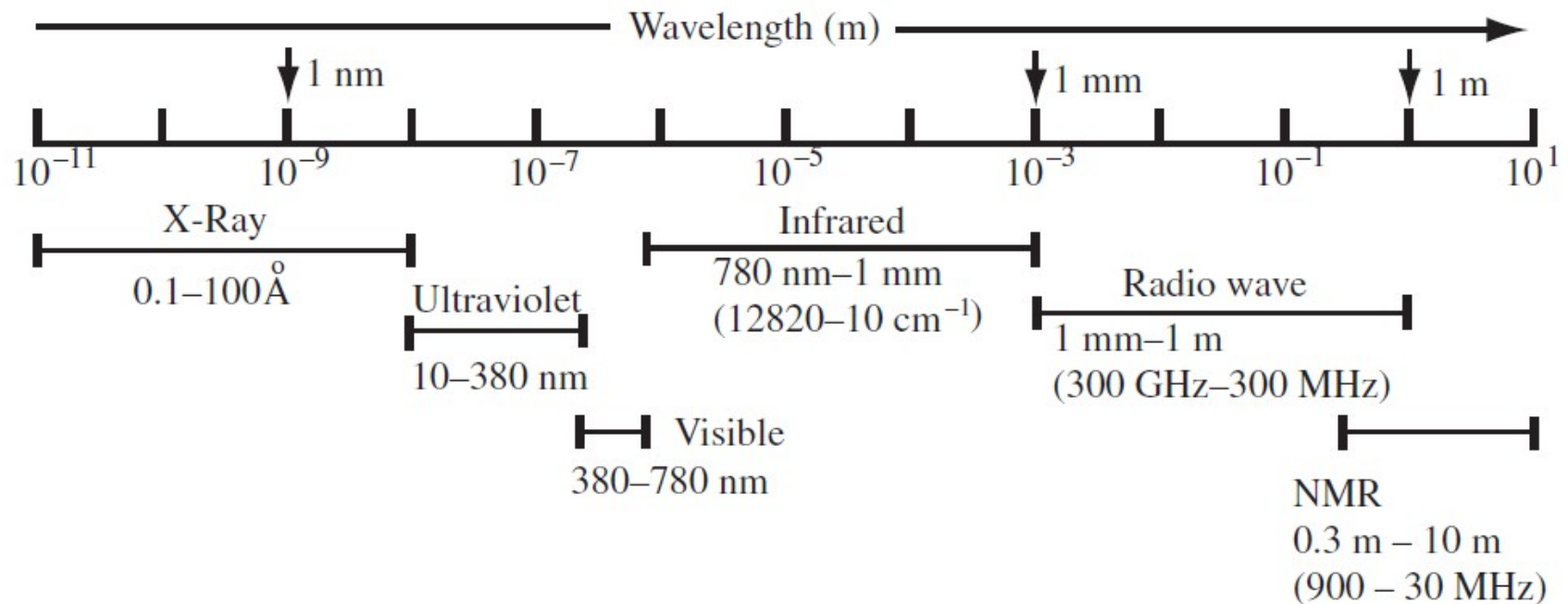
Subject studying interaction of electromagnetic radiation with chemical substance

$$E = h\nu = \frac{hc}{\lambda}$$

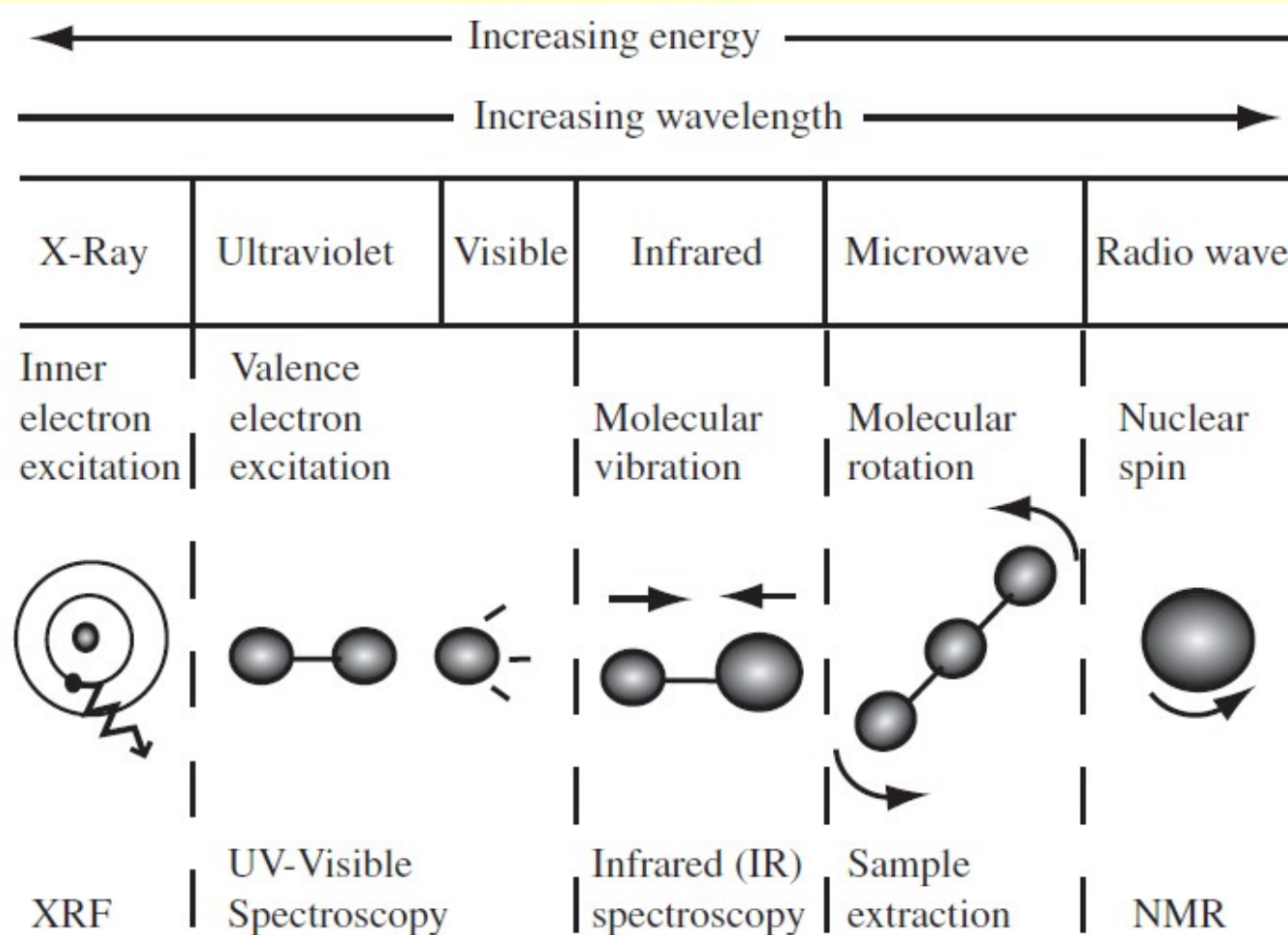
Wavelength



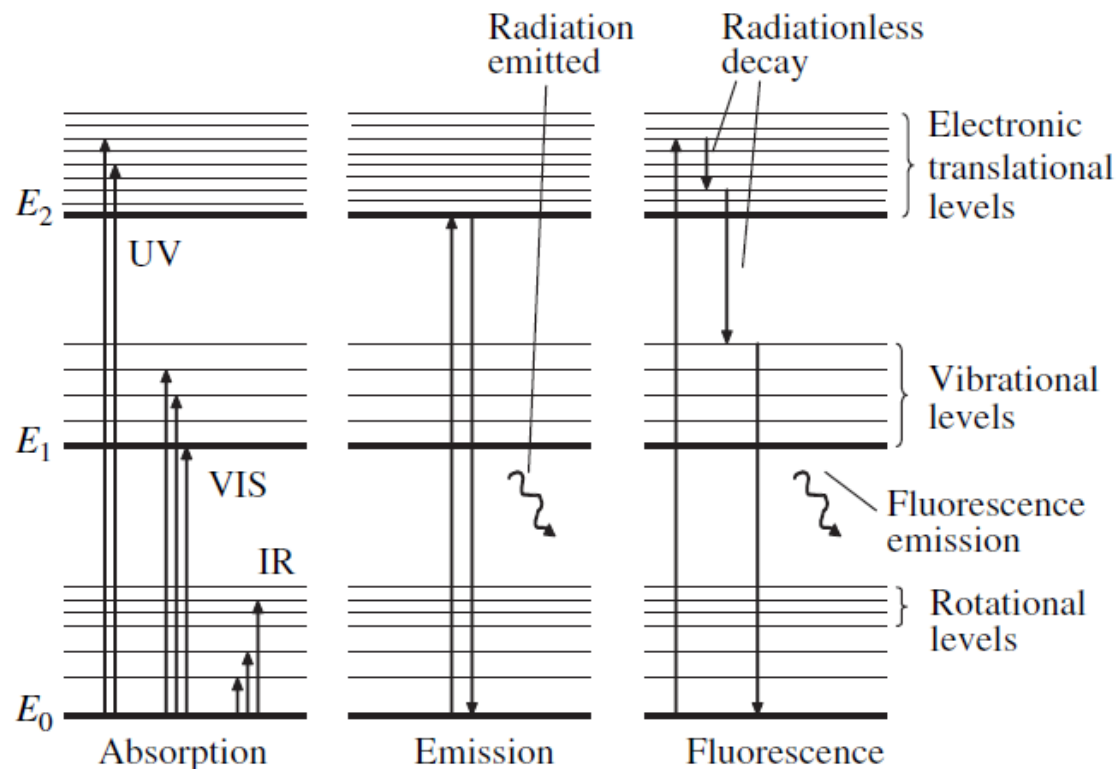
Classification



How the radiation affects molecule



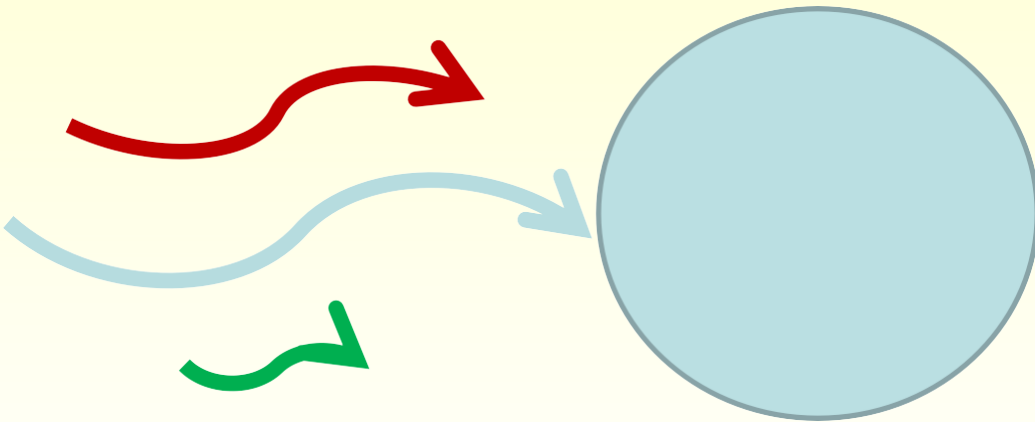
Absorption, emission, fluorescence



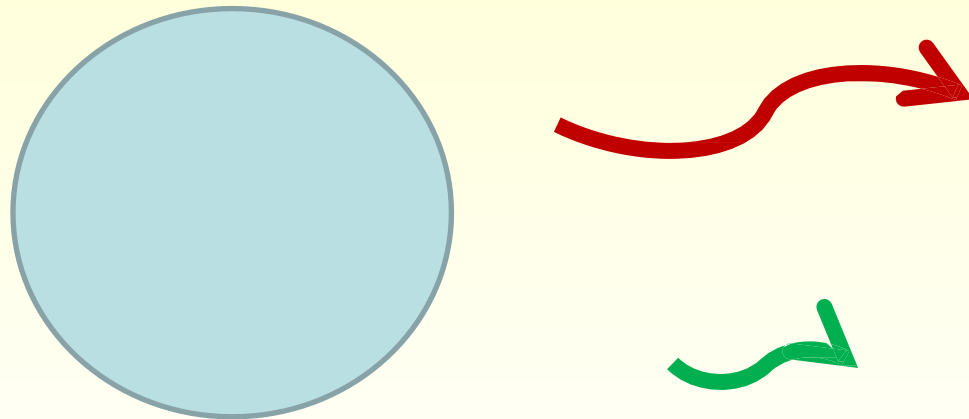
E_0 = Ground level; E_1 , E_2 = Excited states

Energy spacing: vibration > rotation >> translation

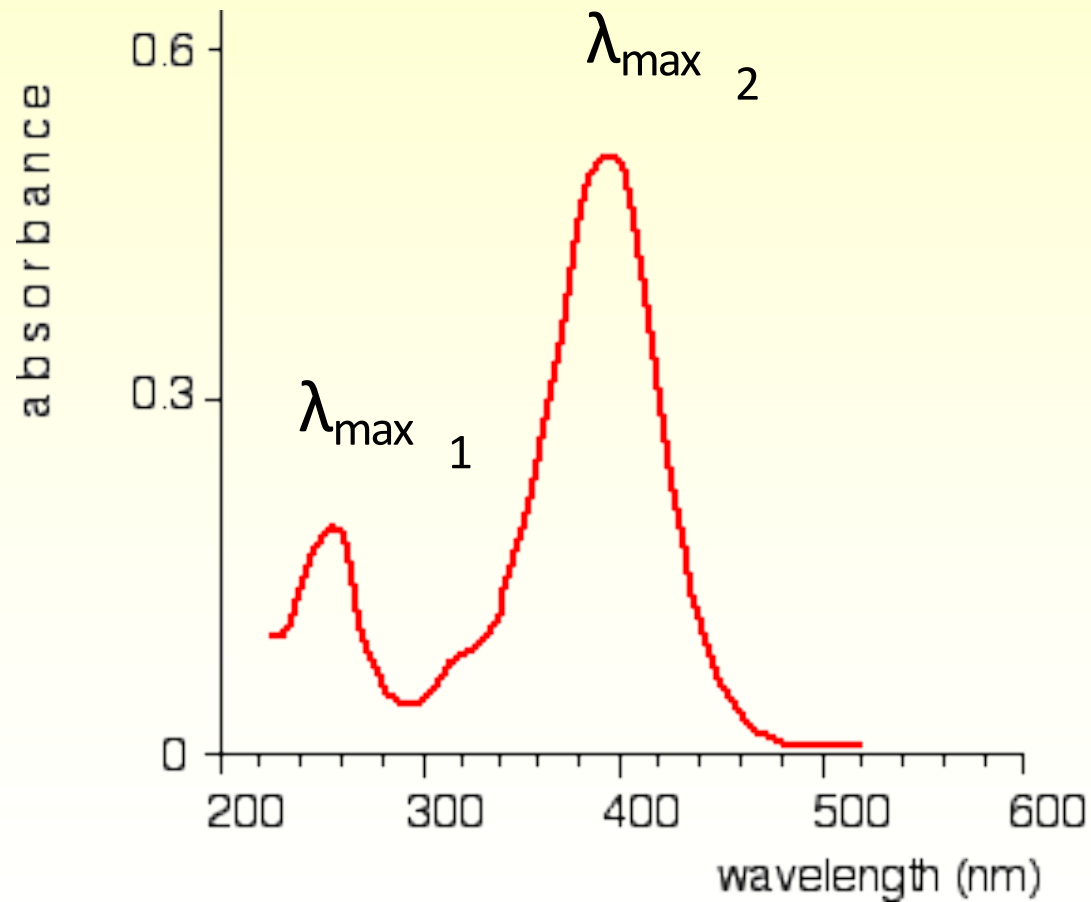
Light absorption



Light absorption



Absorbance spectrum



Transmission

$$T = \frac{P}{P_0}$$

$$T = \frac{P_{\text{sample}}}{P_{\text{blank}}}$$

Absorbance

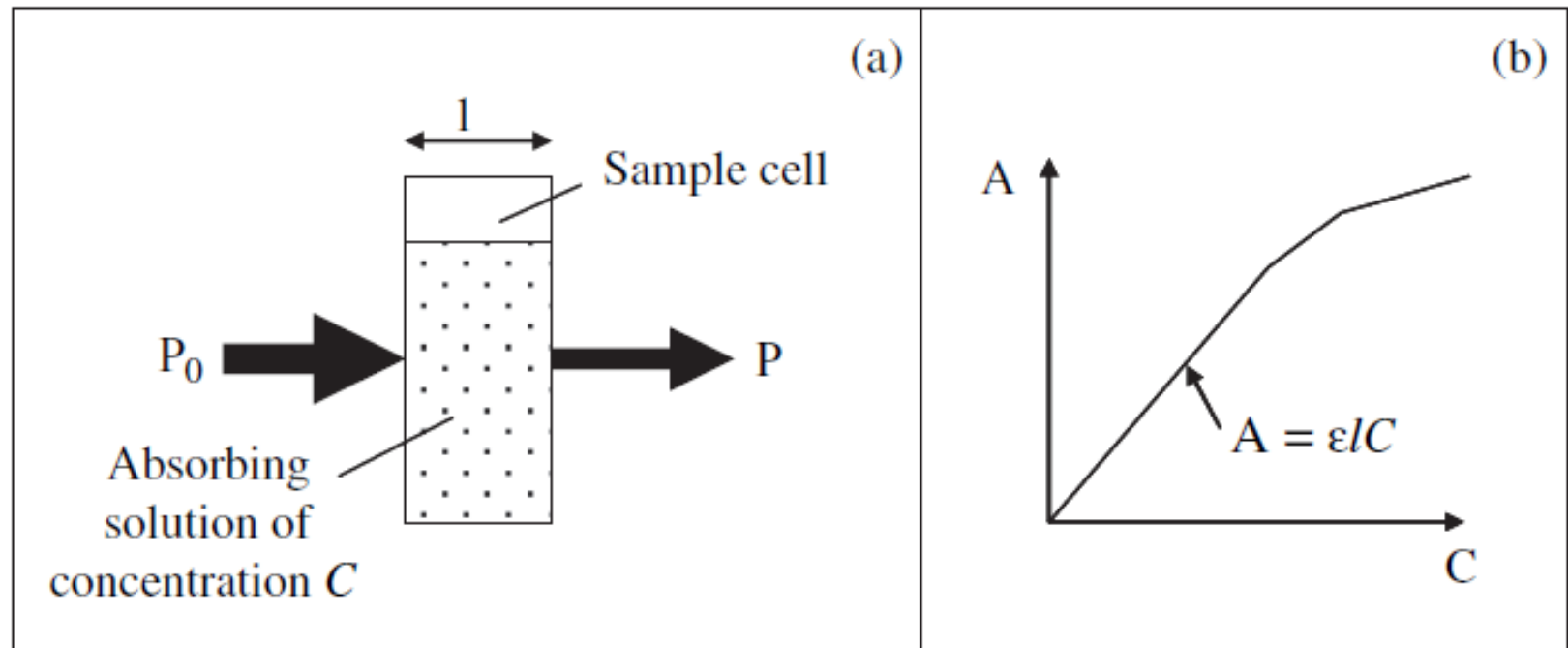
$$A = \log \frac{P_{\text{blank}}}{P_{\text{sample}}} = \log \frac{1}{T}$$

Beer-Lambert Law

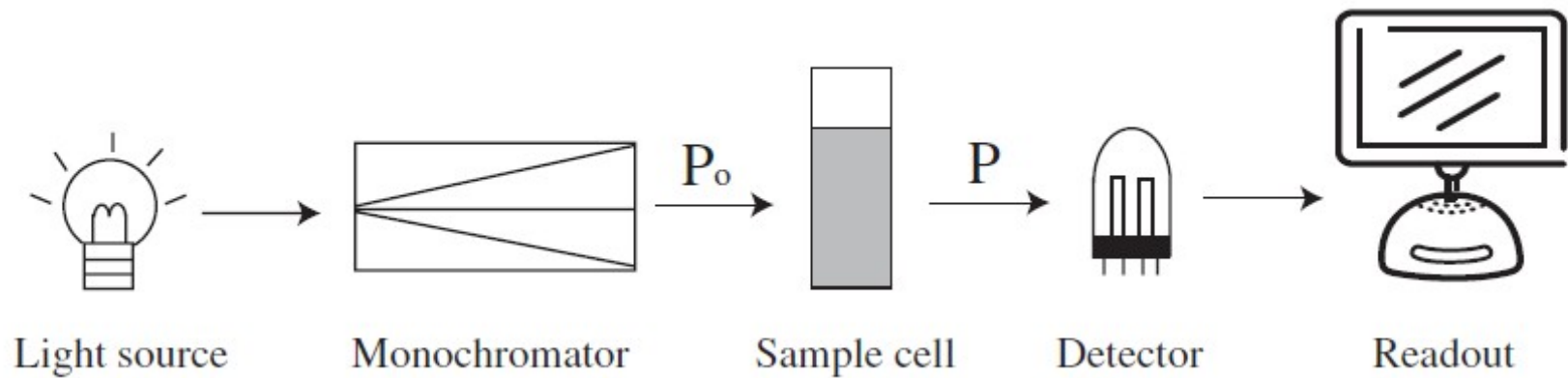
$$A = \epsilon b C$$

b – light path through sample (cuvette width); C –
analyte concentration;
 ϵ – extinction coefficient

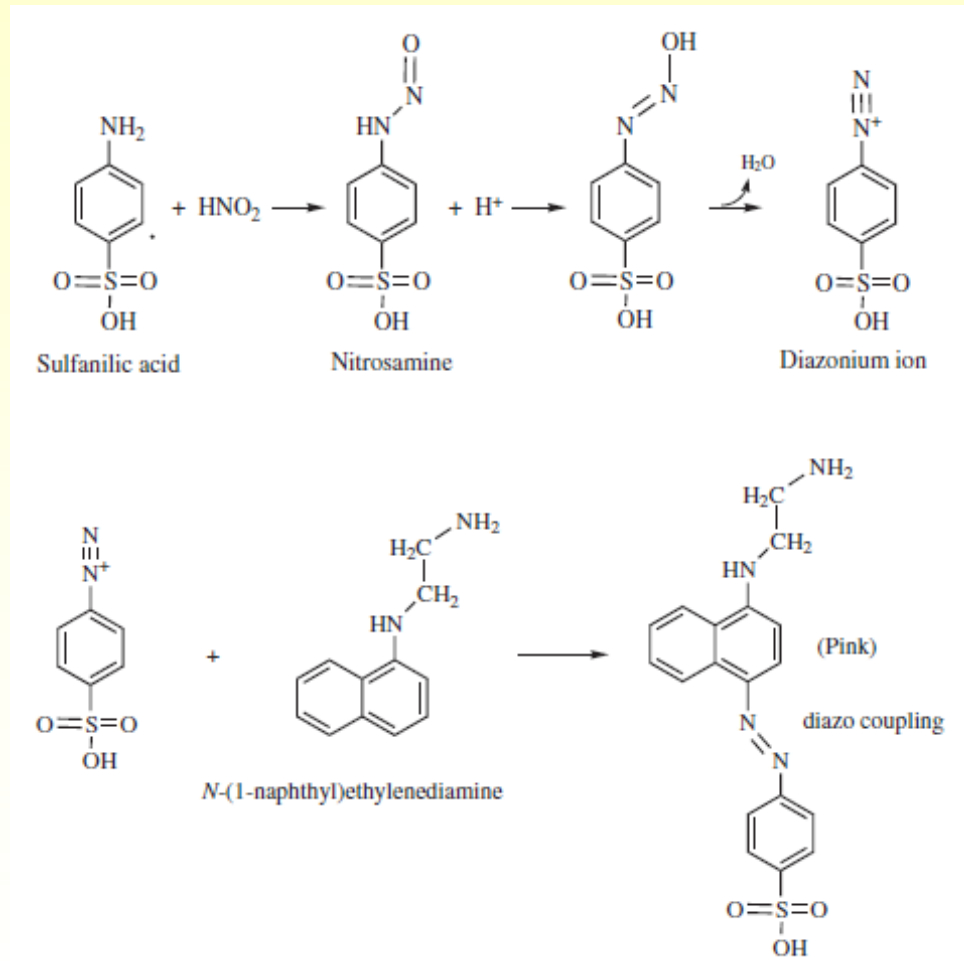
Quantitative analysis by spectroscopic methods



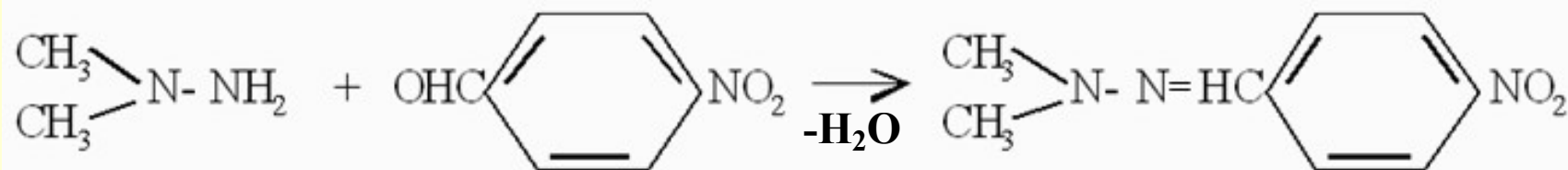
Electron spectroscopy



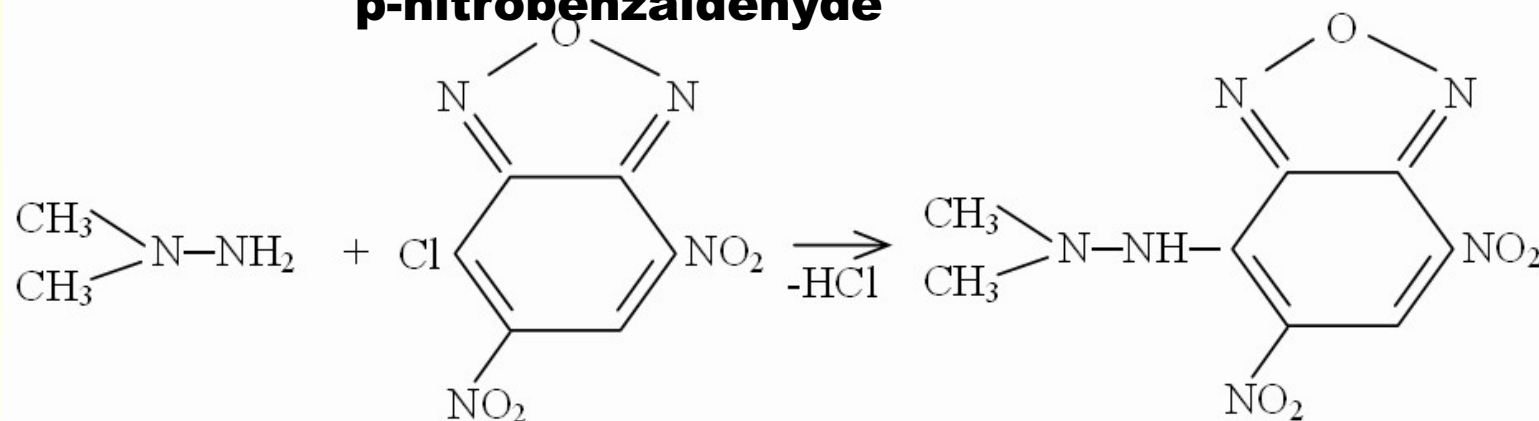
Derivatization: example



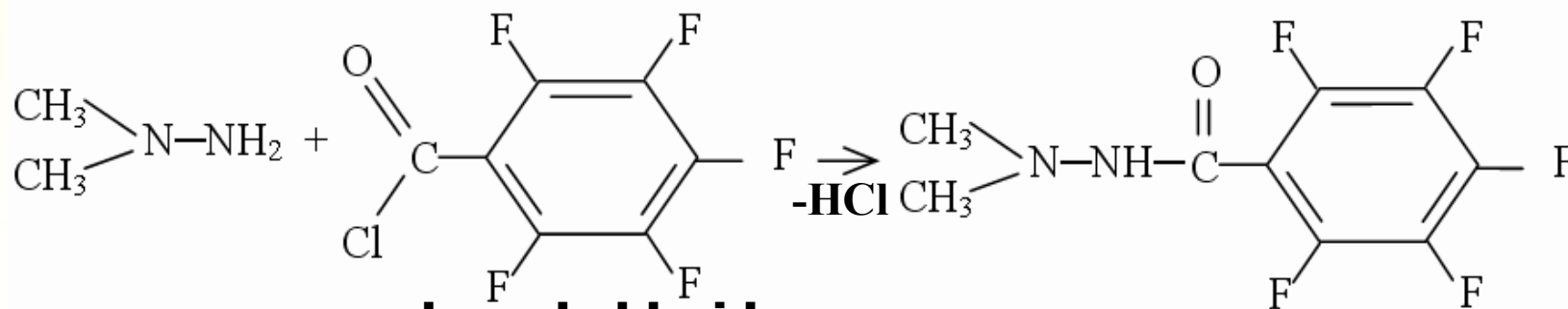
Derivatization of 1,1-dimethylhydrazine



p-nitrobenzaldehyde



5,7-dinitrobenzofurazane



dancyl chloride

Advantages of electron spectroscopy

Simple and inexpensive equipment

Colored substances may be detected visually (by eye)

Many derivatization reagents available

Many sensors and field equipment is based on this method

Disadvantages

Poor selectivity

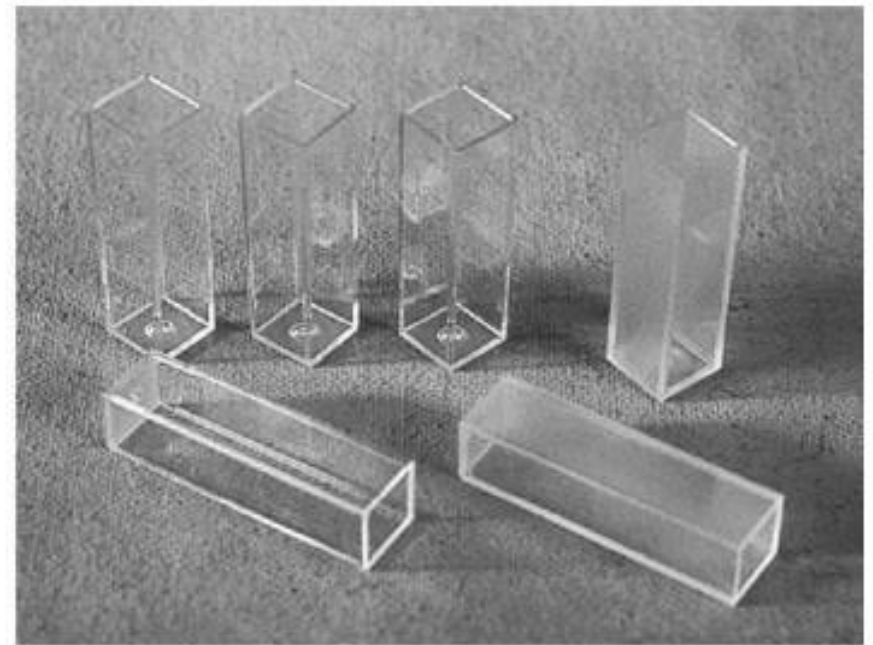
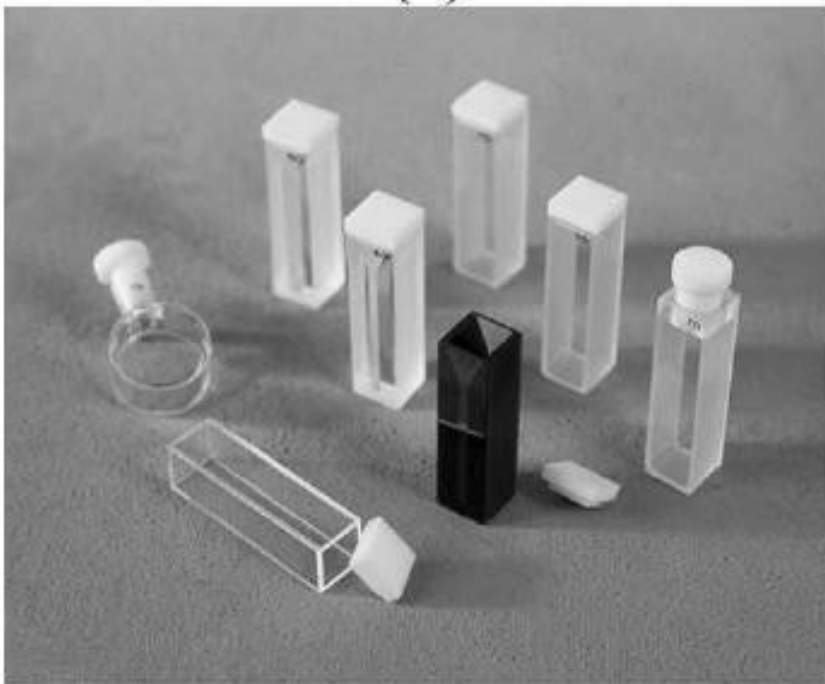
Complex sample preparation when derivatization is needed

Low detection limits are difficult/impossible to achieve

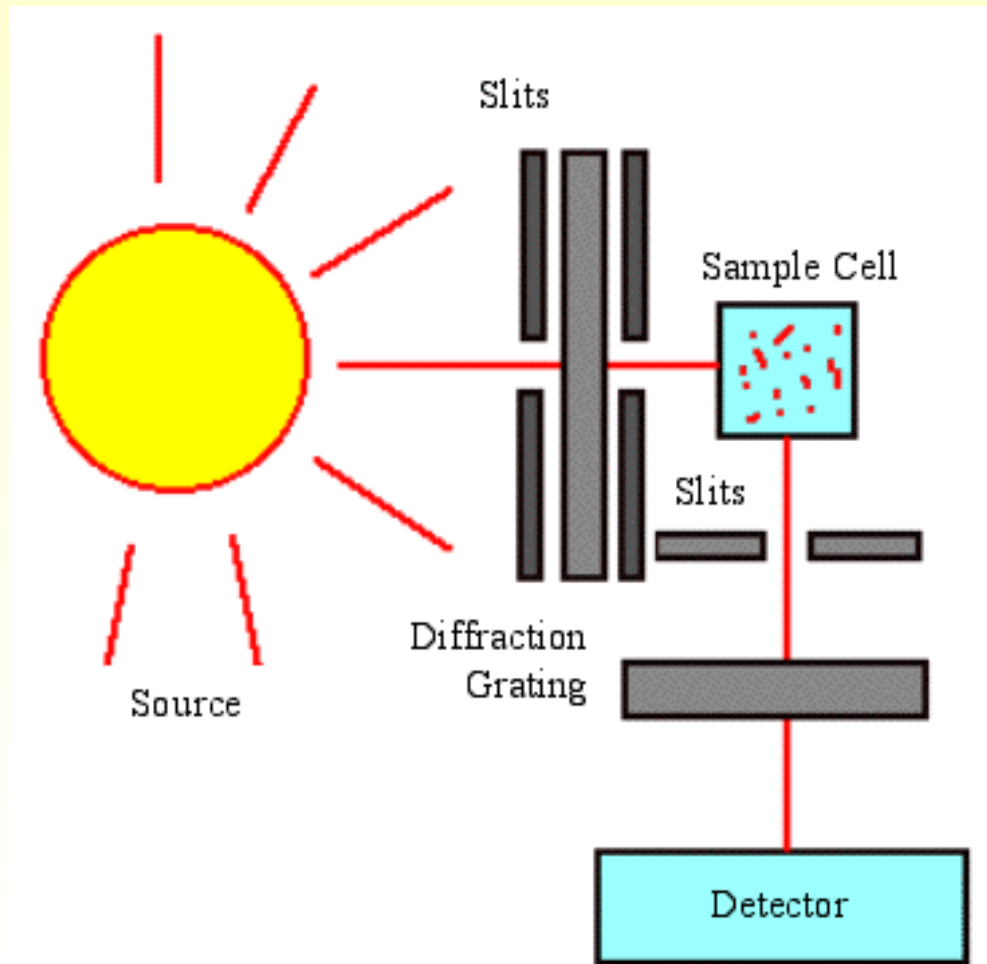
Cuvettes for electron spectroscopy

Quartz – for UV and Visual range

Glass – for Visual range only



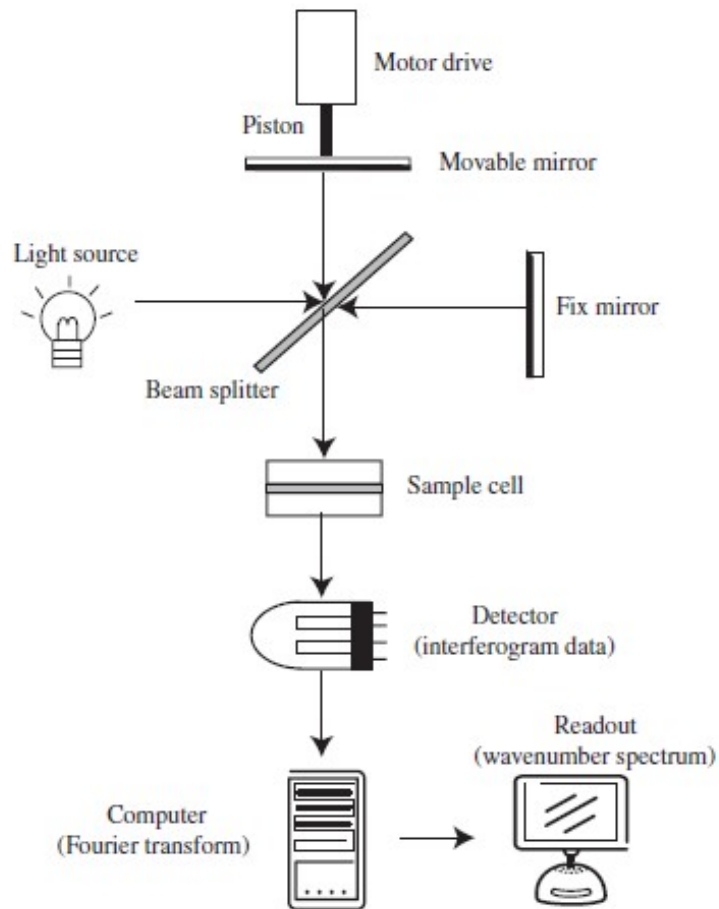
Fluorescence spectroscopy



Very sensitive and
selective method

Popular in biology:
fluorescence markers

Infrared spectroscopy



Ideal for fast identification of pure compounds

Poor sensitivity and selectivity:
not suitable for complex mixtures

Infrared spectrum

