Spektrální analytické metody

Analytické metody

„bulk“ (průměrná složení) vs lokální analýza

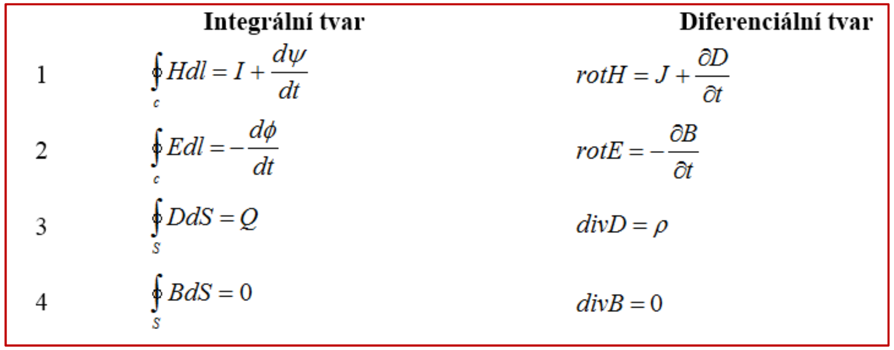
Destruktivní
Semidestruktivní
Nedestruktivní

Atomová spektrometrie
Molekulová spektrometrie

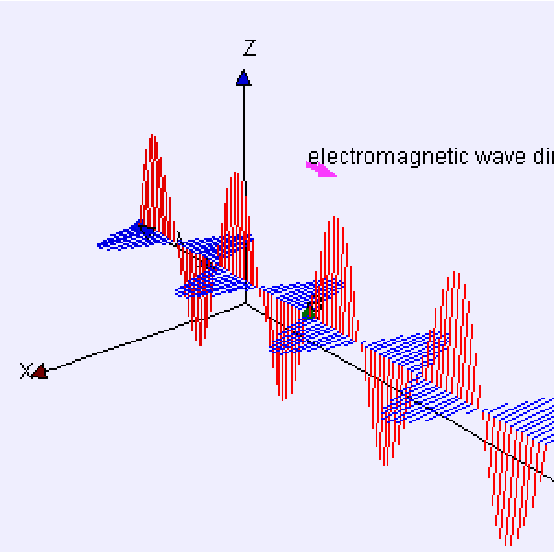
Kvalitativní
Semikvantitativní
Kvantitativní

Elektromagnetické vlnění

Elektromagnetické vlnění (též elektromagnetické záření) je děj, při němž se prostorem šíří příčné vlnění elektrického a magnetického pole. Popsáno je pomocí tzv. Maxwellových rovnic.

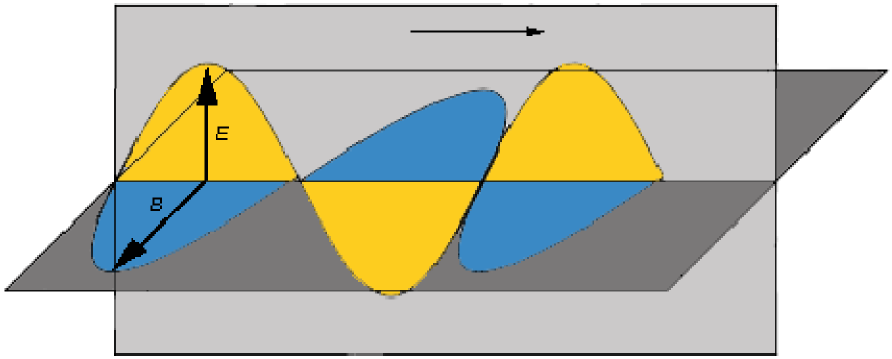


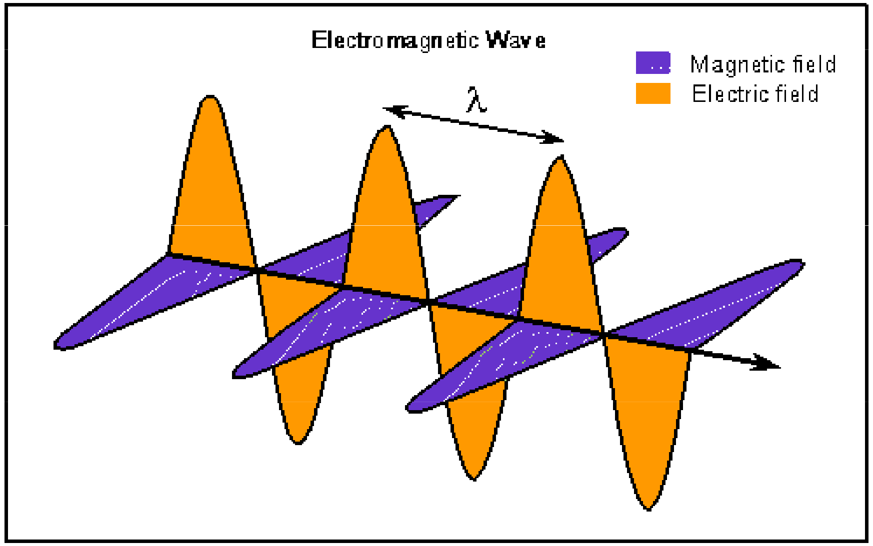
	Integrální tvar	Diferenciální tvar
1	$\oint_c Hdl = I + \frac{d\psi}{dt}$	$rotH = J + \frac{\partial D}{\partial t}$
2	$\oint_c Edl = -\frac{d\phi}{dt}$	$rotE = -\frac{\partial B}{\partial t}$
3	$\oint_S DdS = Q$	$divD = \rho$
4	$\oint_S BdS = 0$	$divB = 0$



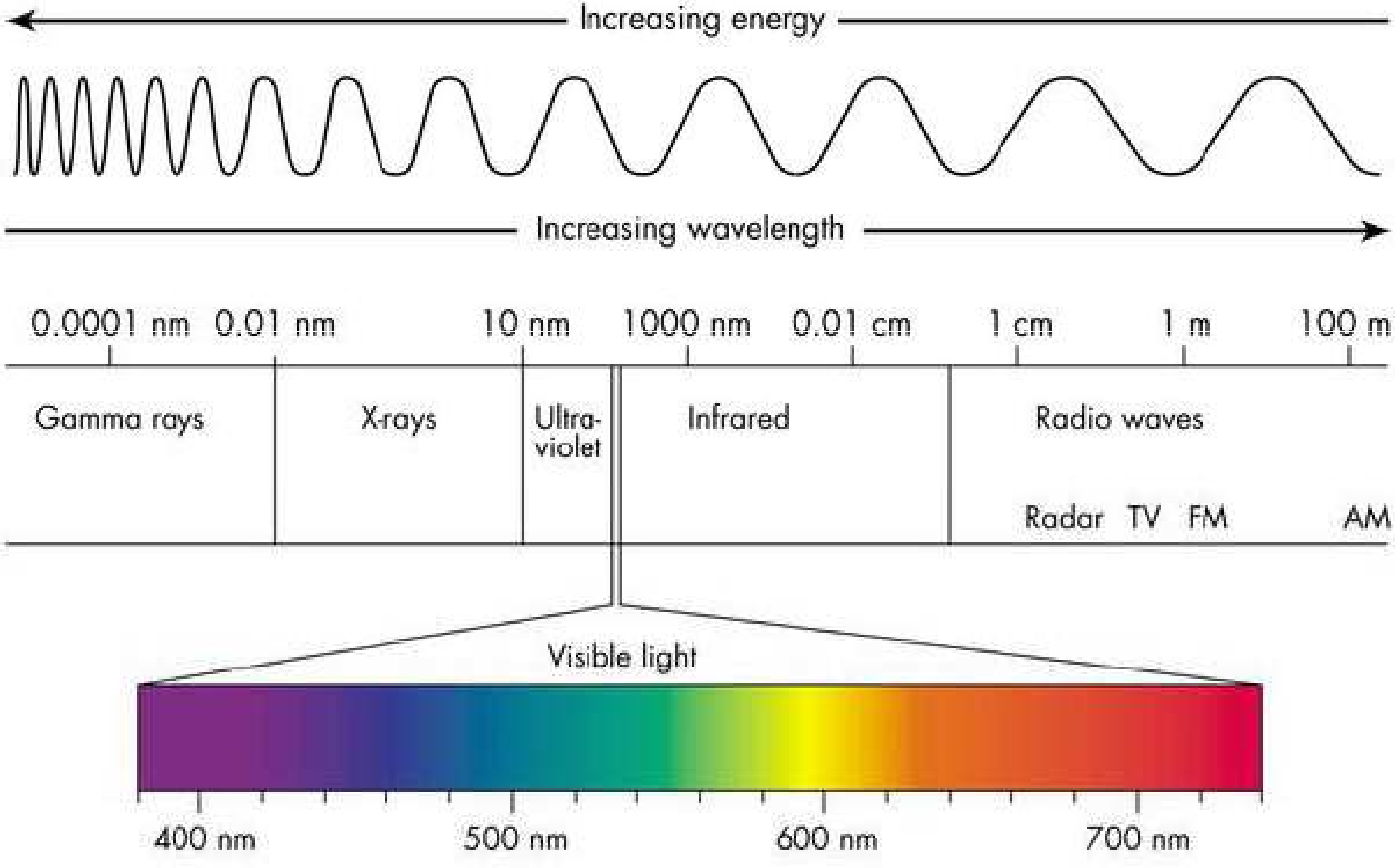
Zahrnuje dvě složky, které jsou na sebe kolmé:

- Intenzita elektrického pole E
- Magnetická indukce B



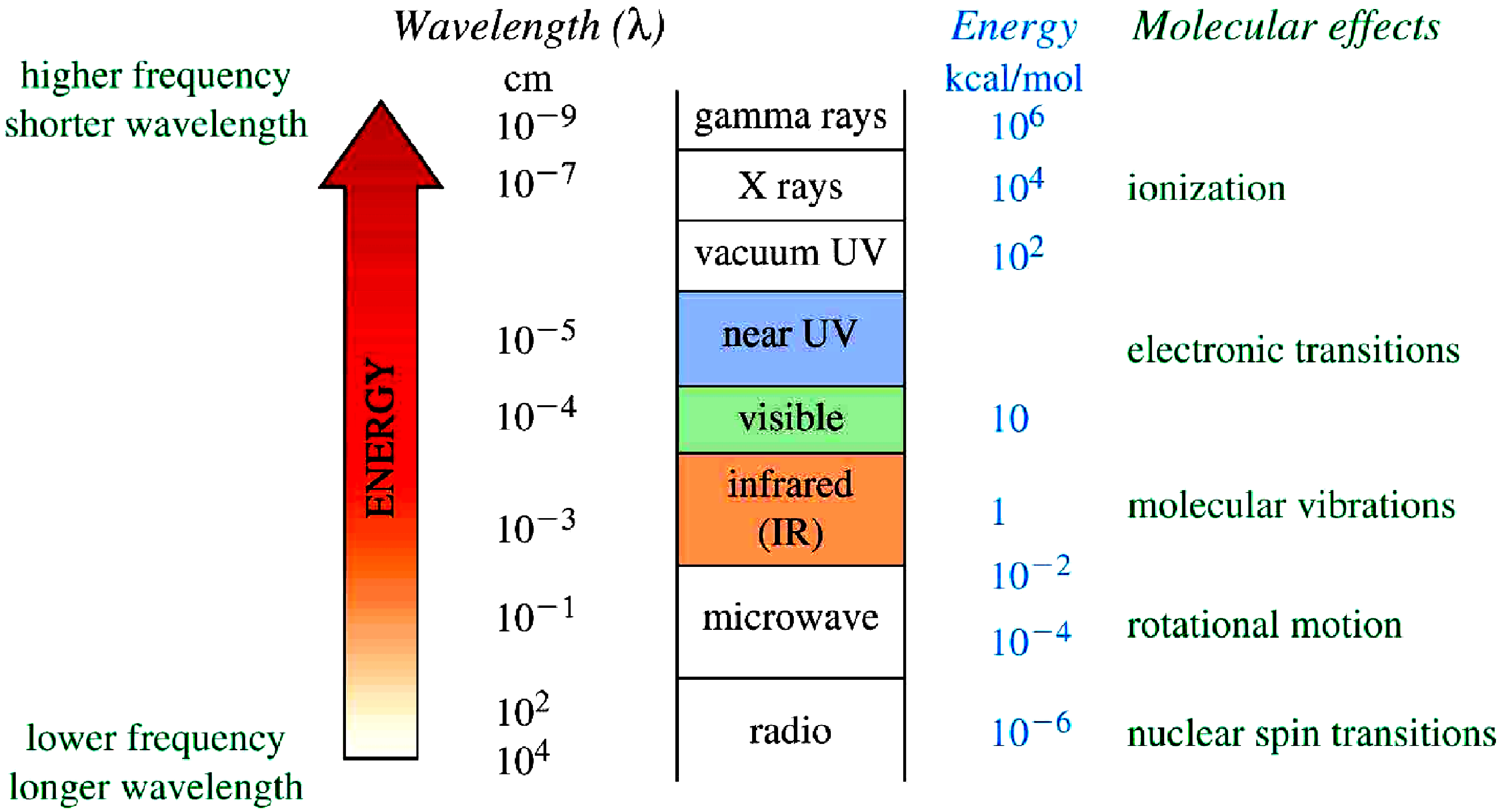


Elektromagnetické záření



Elektromagnetické záření, vlnění	Vlnová délka λ	Použití, výskyt	Pozn.
Radiové vlny Dlouhé (DV) Střední (SV) Krátké (KV) Velmi krátké (VKV) Ultra krátké (UKV)	 2 000 m – 1 000 m 600 m – 150 m 50 m – 15 m 15 m – 1 m 1 m – 0,1 m	Rozhlas, televize	Neionizující záření
Mikrovlny	0,1 m – 0,3 mm	mobilní telefony , GPS, WiMax, Wifi, mikrovlnné trouby, radar	
Infračervené záření	0,3mm – 750 nm	dálkové ovladače, noční vidění, tepelné záření	
Světlo červené oranžové žluté zelené modré fialové 	760 nm – 390 nm	Viditelné světlo	
Ultrafialové záření	390 nm – 10 nm	Opalování, solária, sterilizace	
Rentgenové záření	10 nm – 1 pm	lékařská diagnostika, průmyslová diagnostika	Ionizující záření
Záření gama	< 300 pm	ozařování nádorů, kosmické záření, jaderné reaktory	

EFFECT OF ELECTROMAGNETIC RADIATION ON MOLECULES



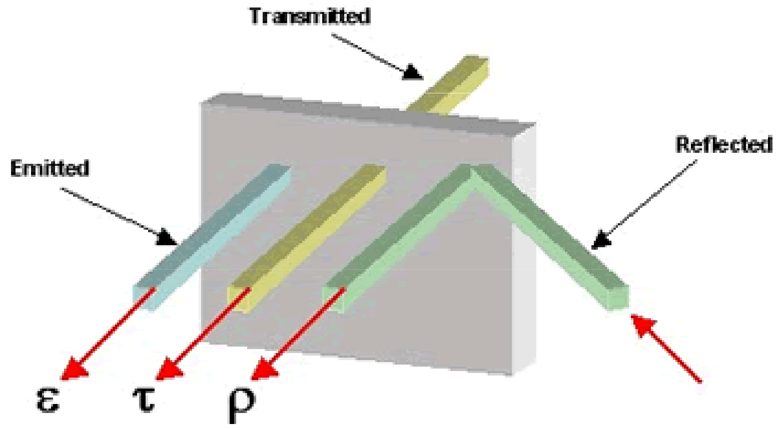
higher frequency
shorter wavelength

Wavelength (λ) cm		*Energy* kcal/mol	*Molecular effects*
10^{-9}	gamma rays	10^{6}	
10^{-7}	X rays	10^{4}	ionization
	vacuum UV	10^{2}	
10^{-5}	near UV		electronic transitions
10^{-4}	visible	10	
10^{-3}	infrared (IR)	1	molecular vibrations
		10^{-2}	
10^{-1}	microwave	10^{-4}	rotational motion
10^{2} 10^{4}	radio	10^{-6}	nuclear spin transitions

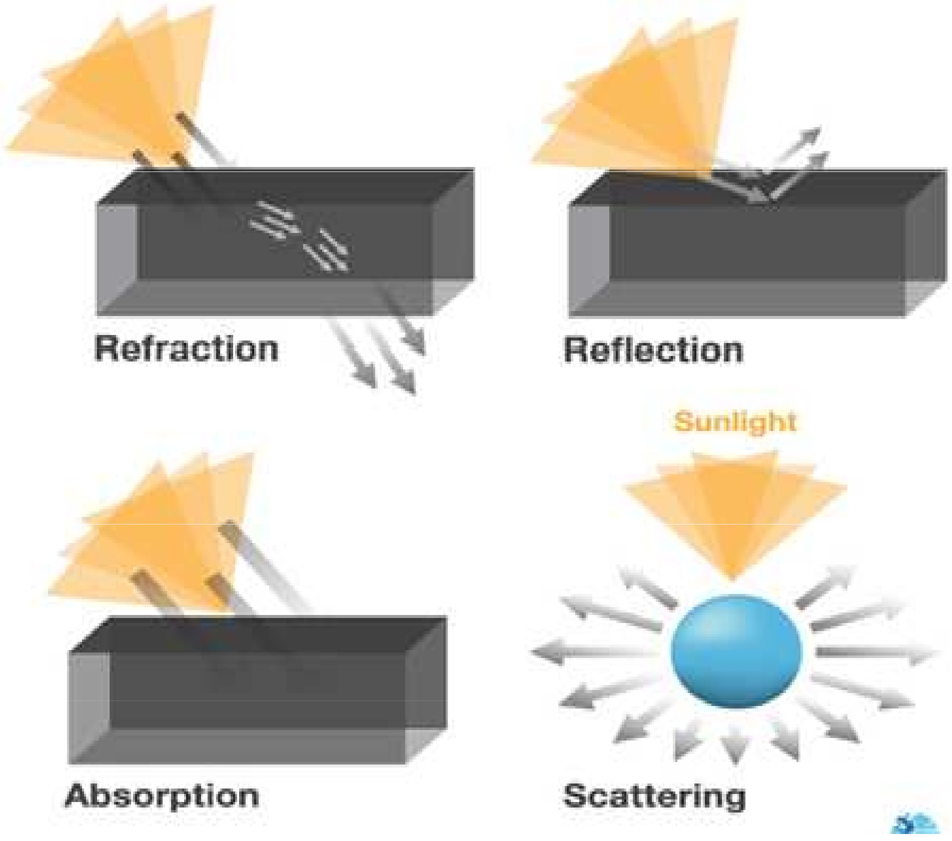
lower frequency
longer wavelength

Graphics source: Wade, Jr., L.G. *Organic Chemistry*, 5th ed. Pearson Education Inc., 2003

Interakce mezi hmotou a energií



$$\varepsilon + \tau + \rho = 1$$



Kvantová teorie

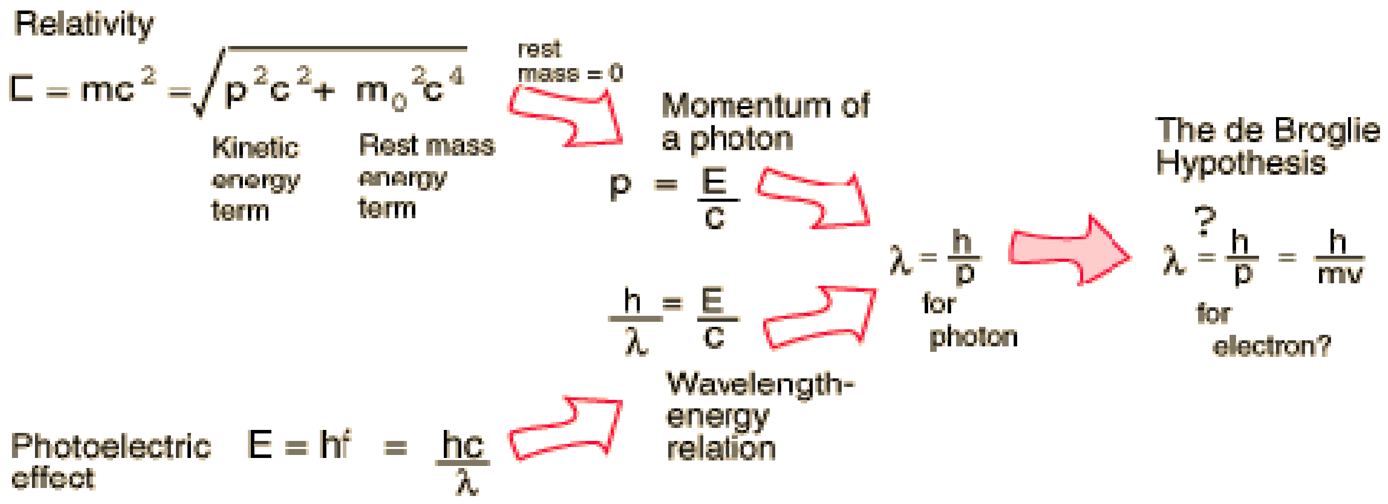
M. Planck: Kvantová teorie $E = h.\nu$

A. Einstein: Fotoelektrický jev

N. Bohr: Kvantový model atomu

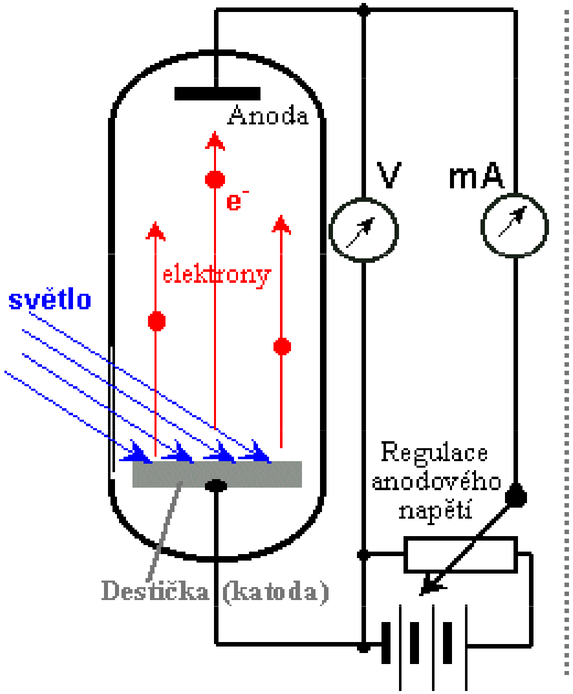
L. De Broglie: Dualita hmota-záření

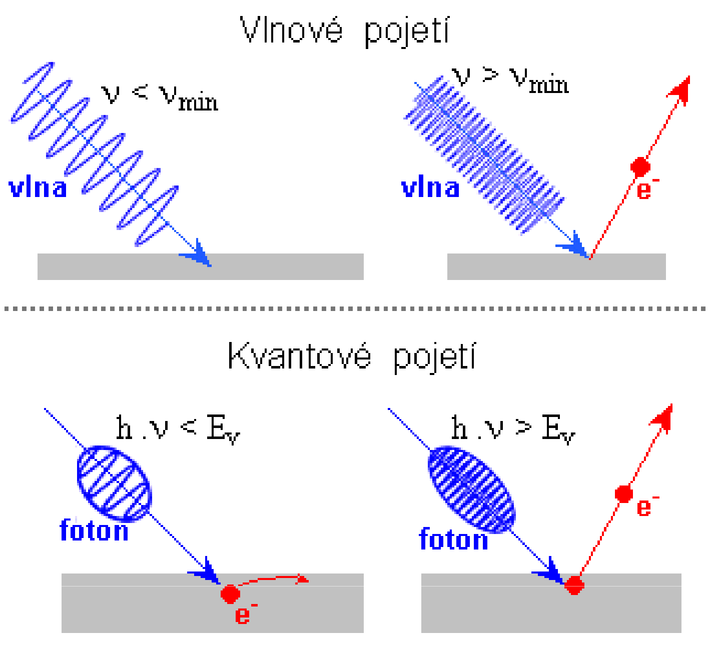
A. Einstein: Speciální teorie relativity $E = m.c^2$



Vlnění

Částice

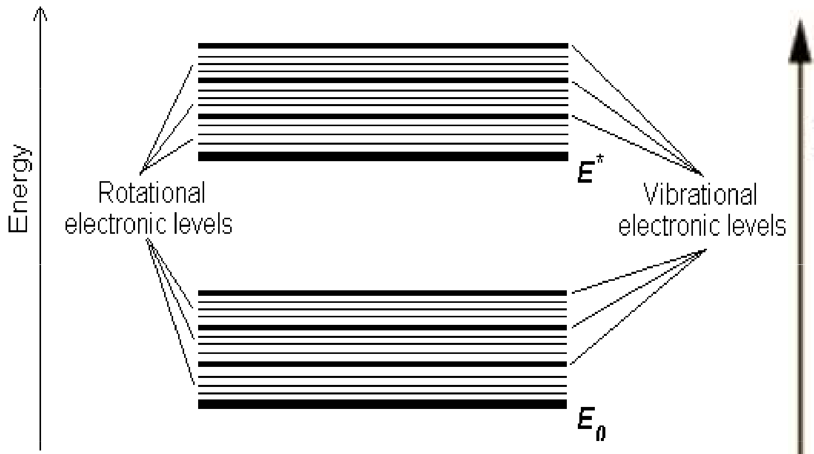


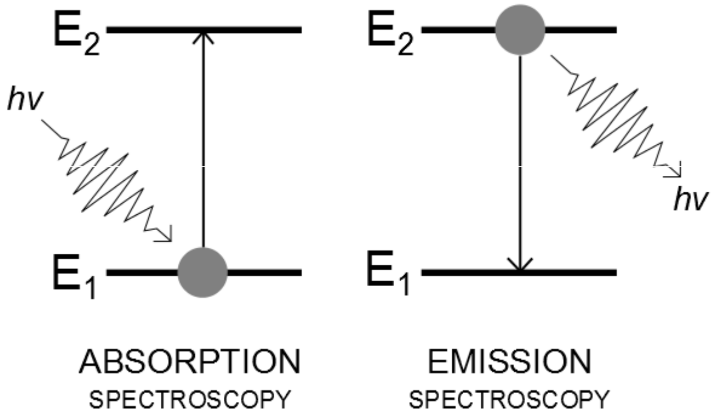


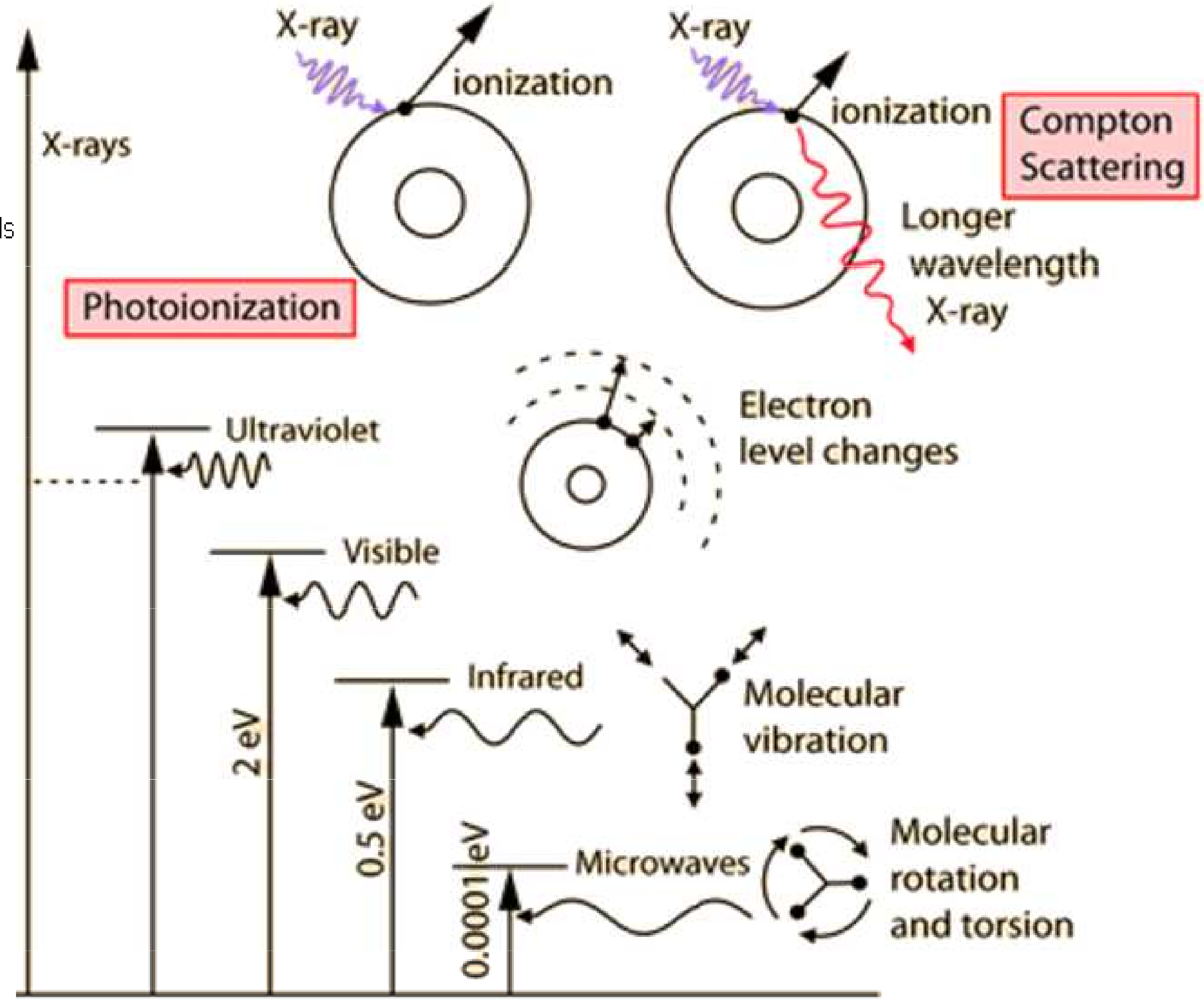
Každá mikročástice o hmotnosti m pohybující se rychlostí v, se může chovat jako vlna o vlnové délce

$$\lambda = \frac{h}{p} = \frac{h}{\gamma m v} = \frac{h}{mv}\sqrt{1 - \frac{v^2}{c^2}}$$

***De Broglieova-Comptonova* vlnová délka**


Energy
Rotational electronic levels
E^*
Vibrational electronic levels
E_0

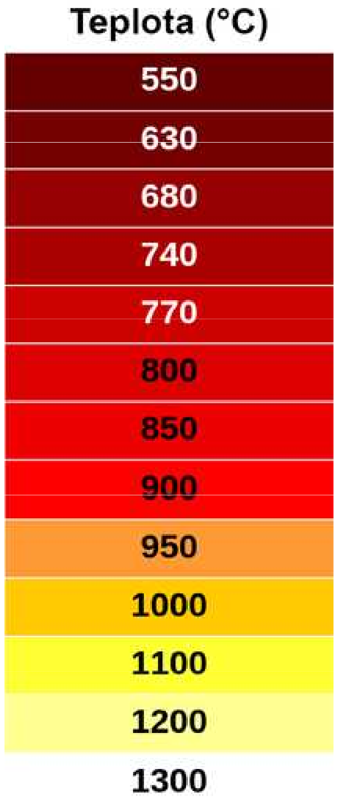

E_2
hv
E_1
ABSORPTION
SPECTROSCOPY
E_2
hv
E_1
EMISSION
SPECTROSCOPY


X-rays
X-ray
ionization
Photoionization
X-ray
ionization
Compton Scattering
Longer wavelength X-ray
Ultraviolet
Electron level changes
Visible
2 eV
Infrared
Molecular vibration
0.5 eV
0.0001eV
Microwaves
Molecular rotation and torsion

Elektromagnetické záření látek

Všechny předměty kolem nás vydávají elektromagnetické záření. To se v případě studených těles nachází v infračervené části spektra, která není pro lidské oko viditelná. S rostoucí teplotou tělesa se vyzařování tepelného záření přesouvá ke kratším vlnovým délkám (k vyšším frekvencím).

Nejnižší teplota, při které je záření daného tělesa pozorovatelné pouhým okem, se označuje jako **Draperův bod** – ten odpovídá zhruba **525 °C**. Při této teplotě vyzařují všechny objekty, bez ohledu na materiál, z něhož jsou vyrobeny, **červené světlo**.



Když předmět **dále zahříváme**, mění se postupně jeho barva z červené přes oranžovou a žlutou k bílé. Při ještě vyšších teplotách se záření posouvá směrem do ultrafialové oblasti. Naše oči ho pak vnímají jako namodralé.

Záření černého tělesa

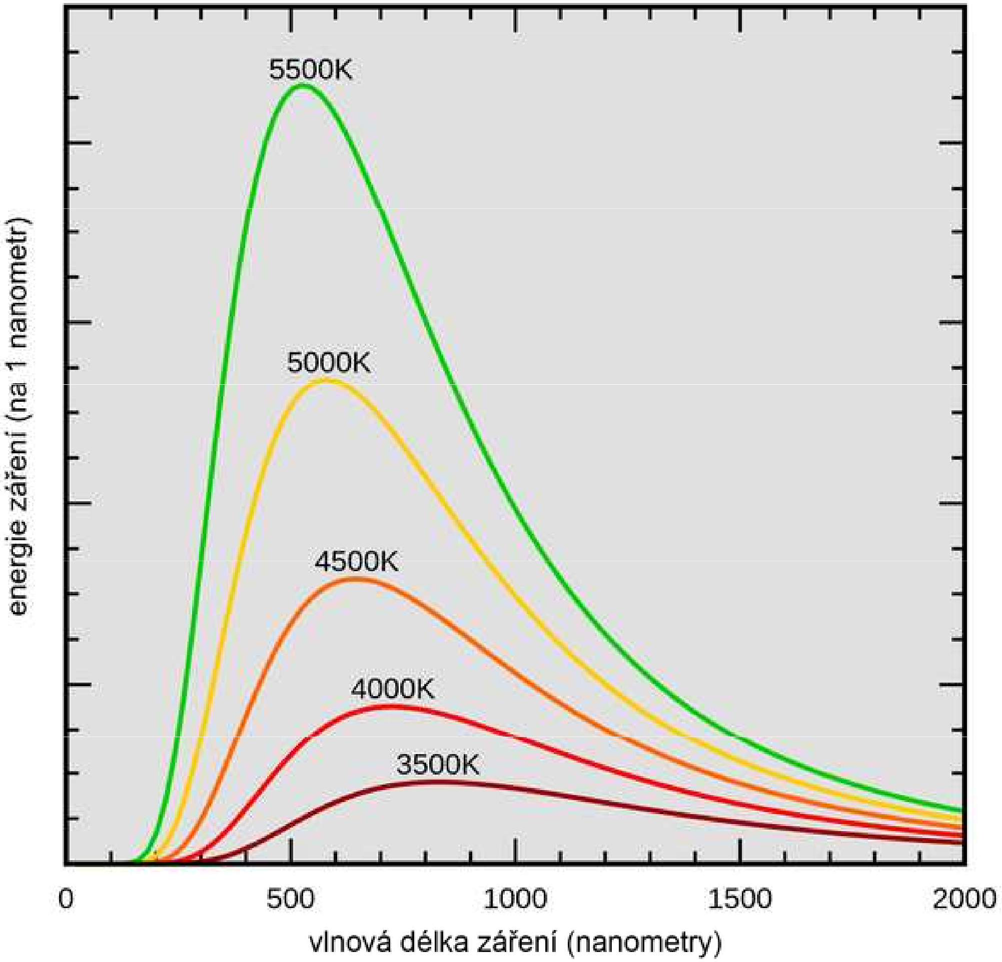
Černé těleso je fyzikální abstrakce tělesa, které dokonale pohlcuje veškerou energii dopadajícího záření. V absolutně černém tělese je v rovnováze vyzařování a pohlcování záření.

Wienův posunovací zákon

S rostoucí teplotou zářiče se posouvá maximální hodnota spektrální hustoty zářivého toku ke kratším vlnovým délkám.

$$\lambda_{\max} = \frac{b}{T}$$

b =2,9.10^{-3} m.K

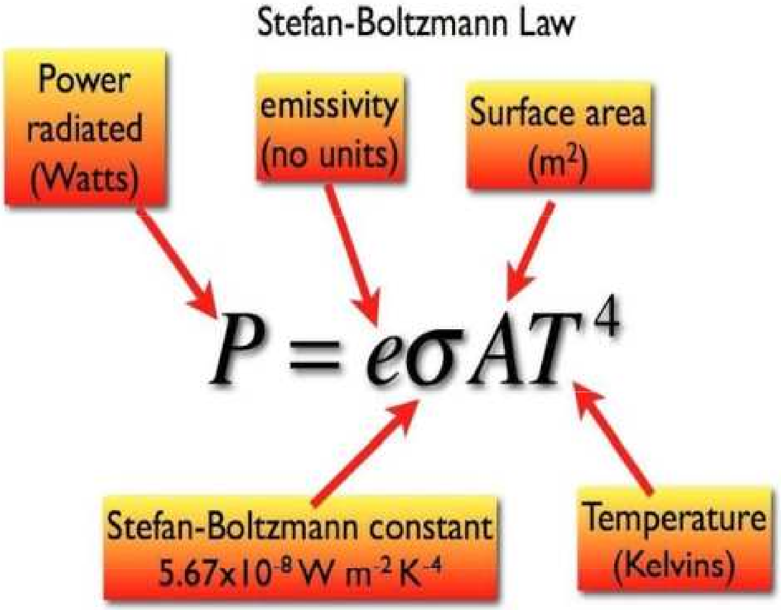


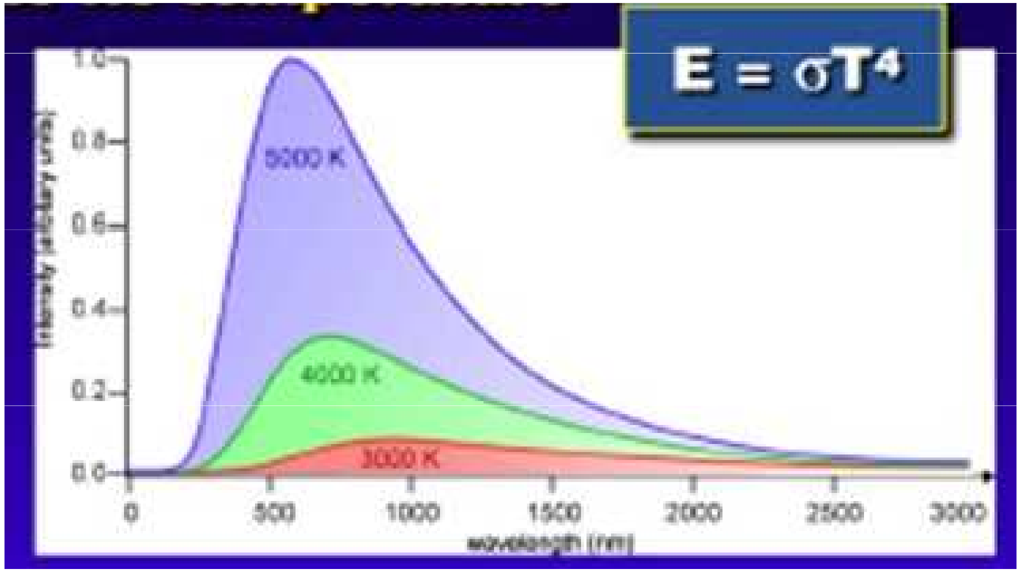
Stefanův-Boltzmannův zákon

Intenzita záření vyzařovaná absolutně černým tělesem roste úměrně čtvrté mocnině termodynamické teploty.

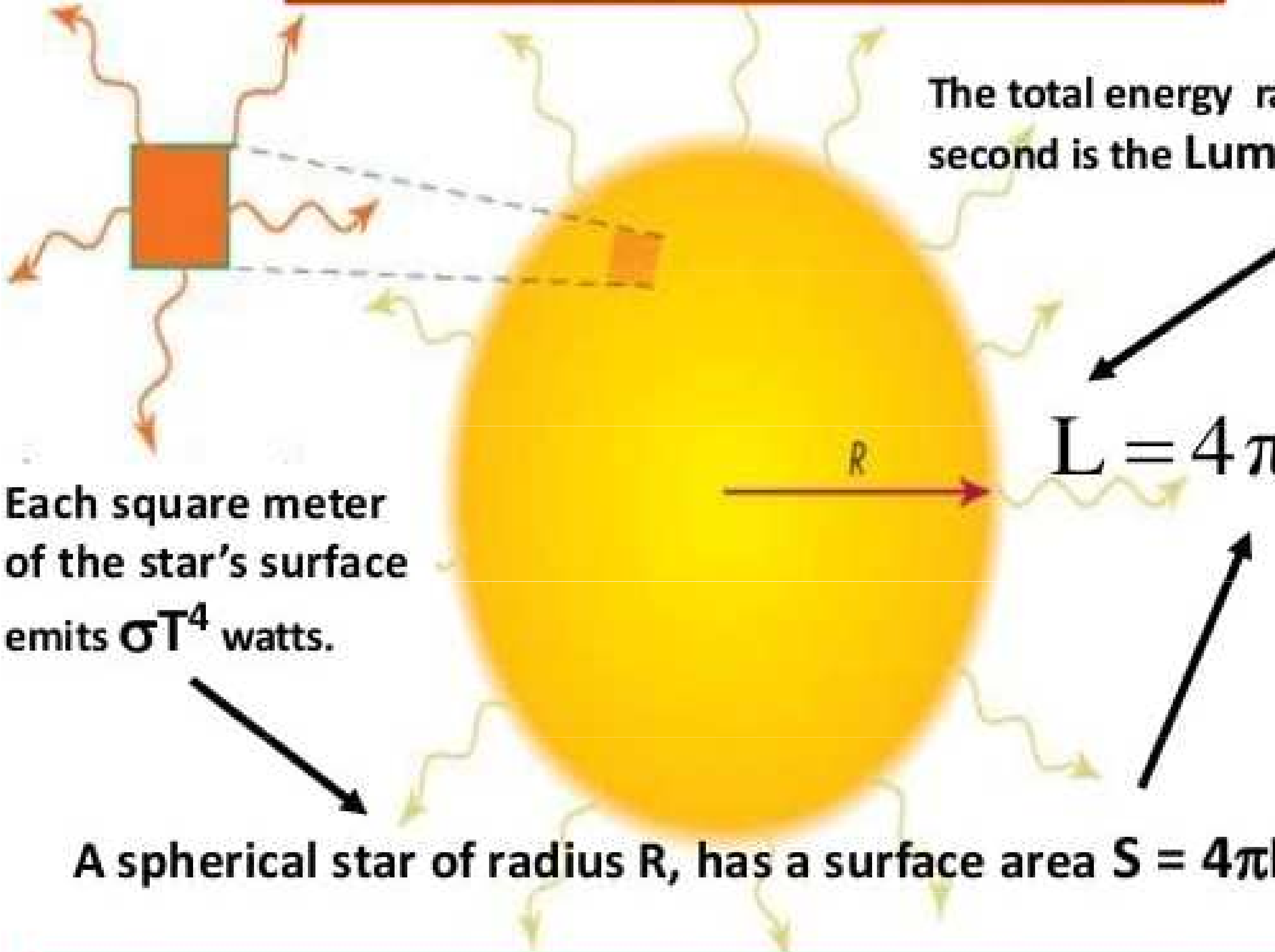
$$I = \sigma T^4$$

I – celková intenzita záření (podíl výkonu a plochy) [$W \cdot m^{-2}$]
σ - Stefan-Boltzmannova konstanta $\sigma = 5{,}67.10^{-8}\ W.m^{-2}\ .K^{-4}$
T - termodynamická teplota





The Stefan-Boltzmann Law



The total energy radiated per second is the **Luminosity L**:

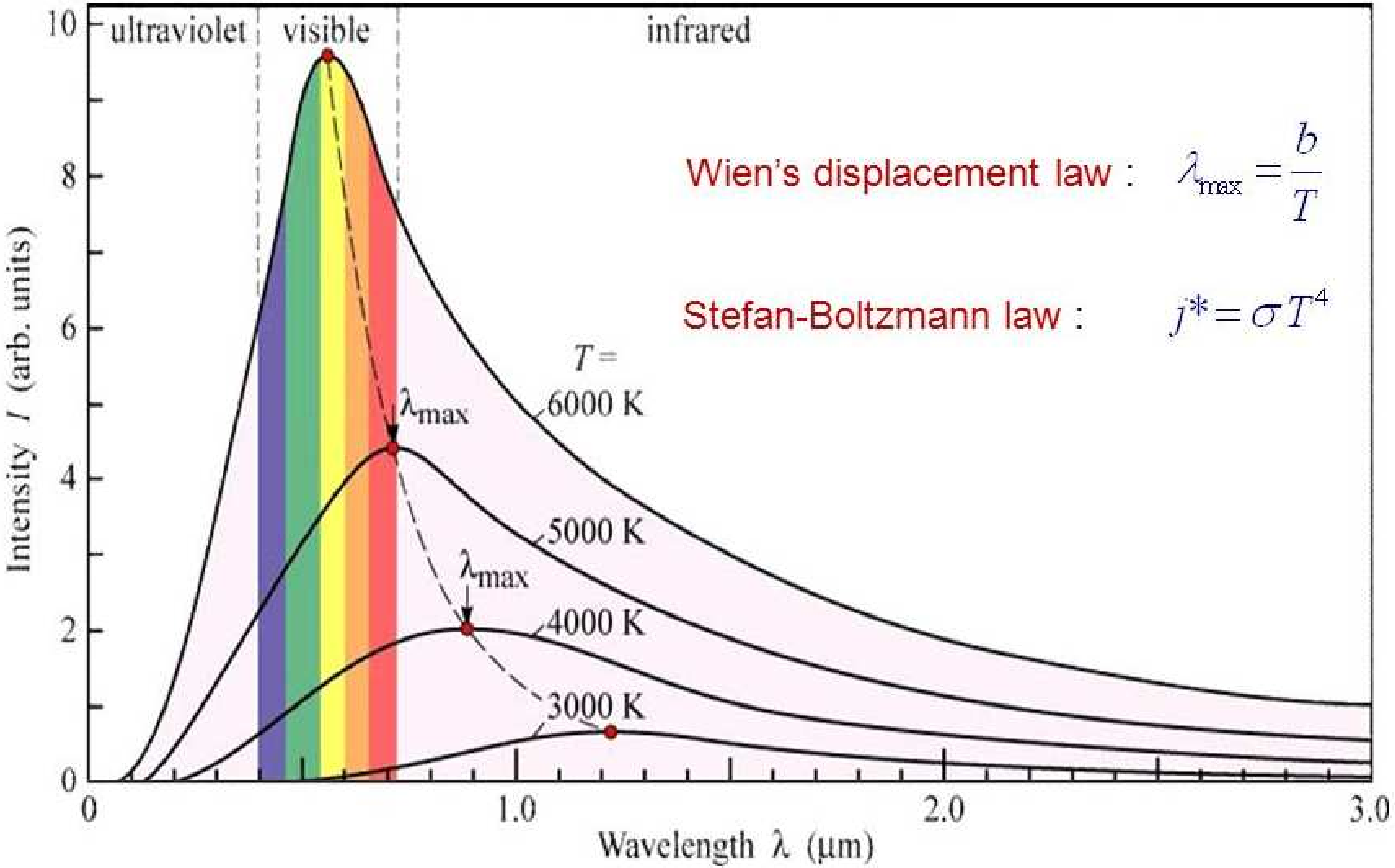
$$L = 4\pi R^2 \sigma T^4$$

Each square meter of the star's surface emits σT^4 watts.

A spherical star of radius R, has a surface area $S = 4\pi R^2$

A star's luminosity is related to both a star's size and temperature:
(a) Hotter stars emit more. (b) Larger stars emit more.

Blackbody Radiation



Wien's displacement law : $\lambda_{\max} = \frac{b}{T}$

Stefan-Boltzmann law : $j^* = \sigma T^4$

Rayleighův-Jeansův zákon

= přibližný zákon pro záření černého tělesa platný pro dlouhé vlnové délky (rádiové vlny a mikrovlny), tj. pro energii fotonu $h.\nu^4 << k.T$.

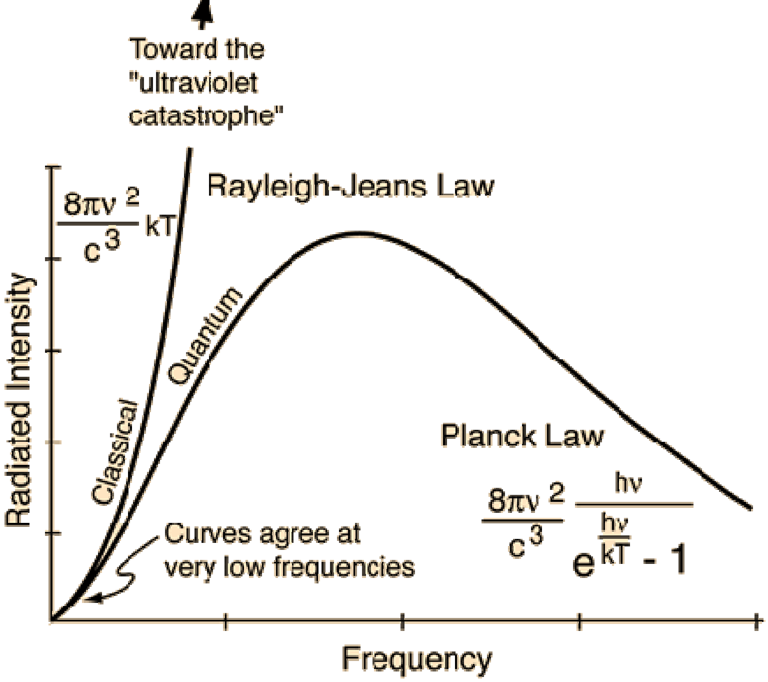
Zářivý výkon připadající na 1 m^2 povrchu černého tělesa a na jednotkový interval vlnové délky, resp. frekvence, je

$$I_\lambda = 2\pi \, c \, k \, T / \lambda^4$$

$$I_\nu = 2\pi \, c \, k \, T \, \nu^2 / c^2$$

λ ... vlnová délka záření,
T ... absolutní teplota zářiče
c ... rychlost světla
k ... Boltzmannova konstanta

Zásadní nesoulad s experimentem se objevuje u Rayleighova-Jeansova zákona od oblasti ultrafialového záření. S klesající vlnovou délkou by spektrální intenzita vyzařování měla růst do nekonečna a absolutně černé těleso by vydávalo tepelné záření o nekonečném výkonu (tzv. **ultrafialová katastrofa**).



Planckův vyzařovací zákon

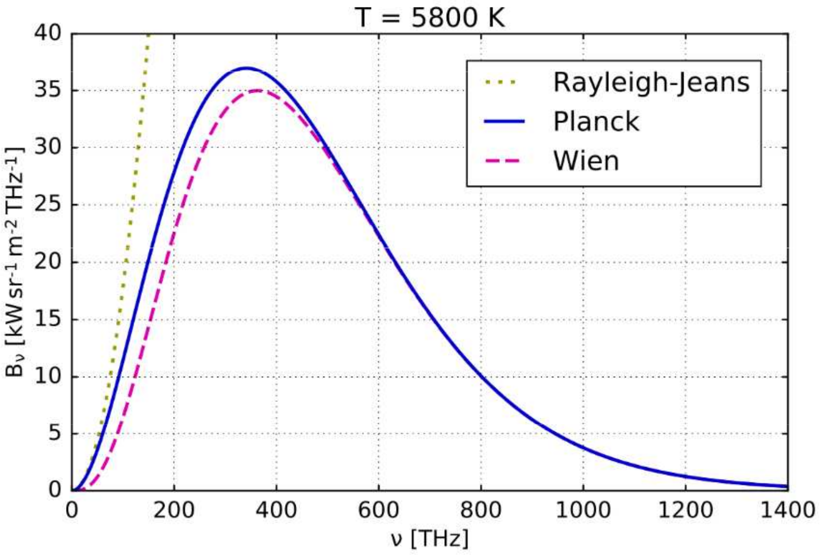
Planckův vyzařovací zákon vyjadřuje závislost intenzity záření absolutně černého tělesa na frekvenci ω.

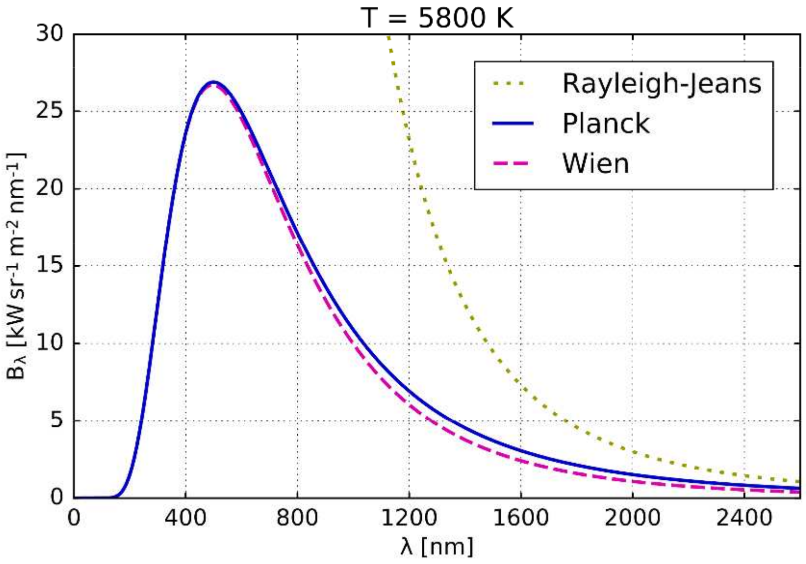
$$u(\nu)\, \mathrm{d}\nu = \frac{8\pi h\nu^3}{c^3} \frac{1}{\left(e^{\frac{h\nu}{kT}} - 1\right)}\, \mathrm{d}\nu$$

Záření o frekvenci f může být vyzařováno, nebo pohlcováno jen po kvantech energie o velikosti e = h . ν

$$B_\nu(\nu, T) = \frac{2h\nu^3}{c^2} \frac{1}{e^{\frac{h\nu}{k_B T}} - 1}$$

$$B_\lambda(\lambda, T) = \frac{2hc^2}{\lambda^5} \frac{1}{e^{\frac{h\nu}{\lambda k_B T}} - 1}$$





Spektrografie

Semikvantitativní

Semidestruktivní

Jiskra
El. oblouk

Emisní spektra se zachycují na fotografickou desku

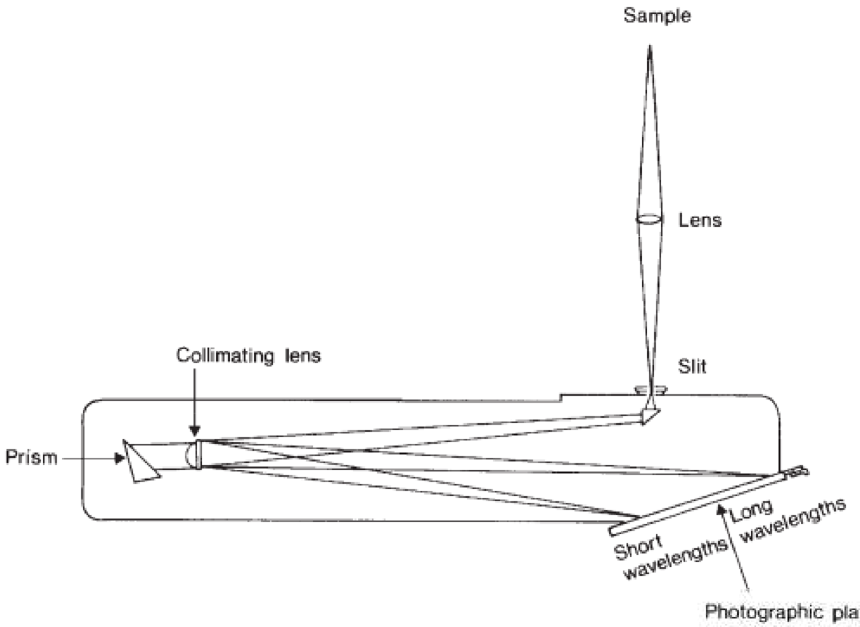
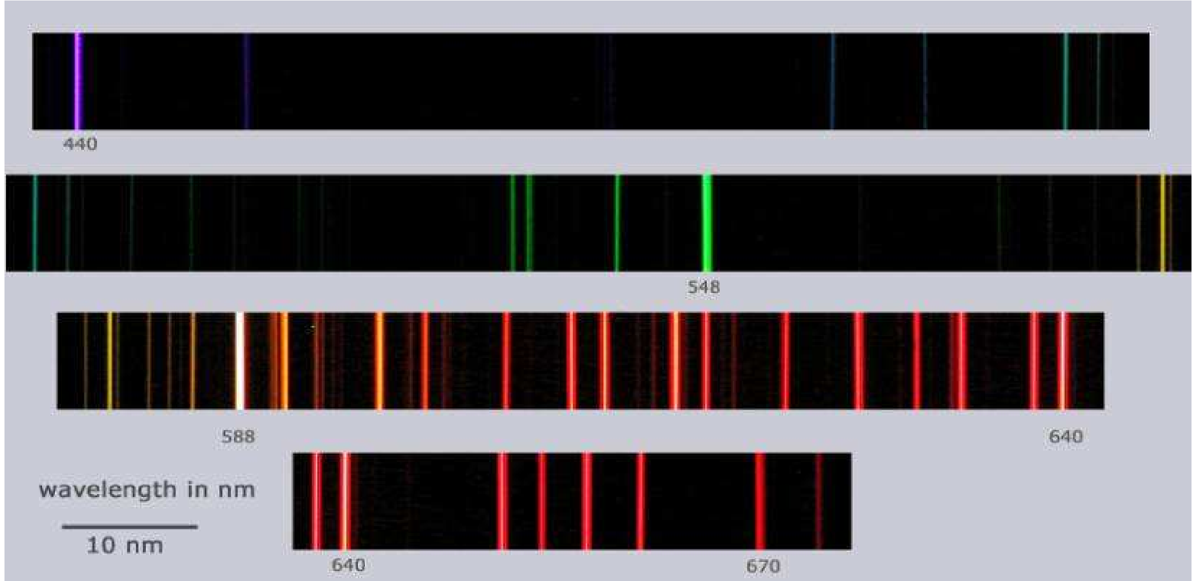


Figure 2.2 Schematic drawing of an optical emission spectrograph. Light from the sample is focused onto the input slit of the spectrograph and is then dispersed via a prism (or diffraction grating) and recorded on a photographic plate. (Adapted from Britton and Richards, 1969; Fig. 108, by permission of Thames and Hudson Ltd.)

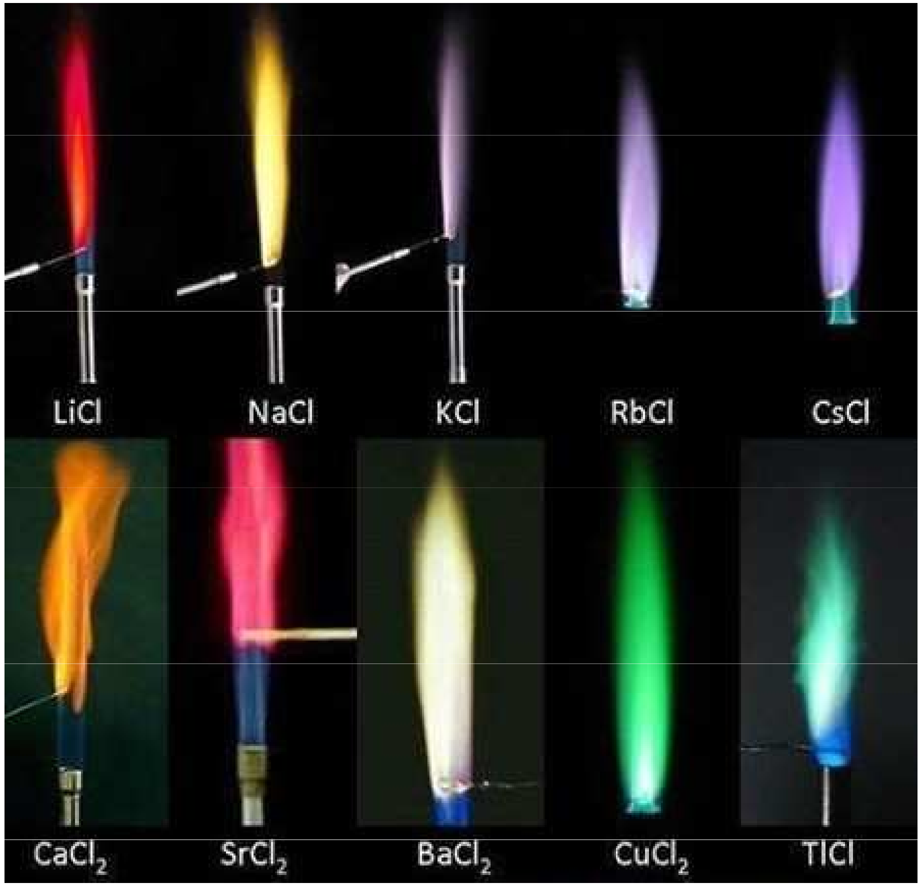


Emission Spectra of the Elements

field·tested·systems™

REAL-TIME CLASSROOM SPECTROSCOPY

Plamenový test

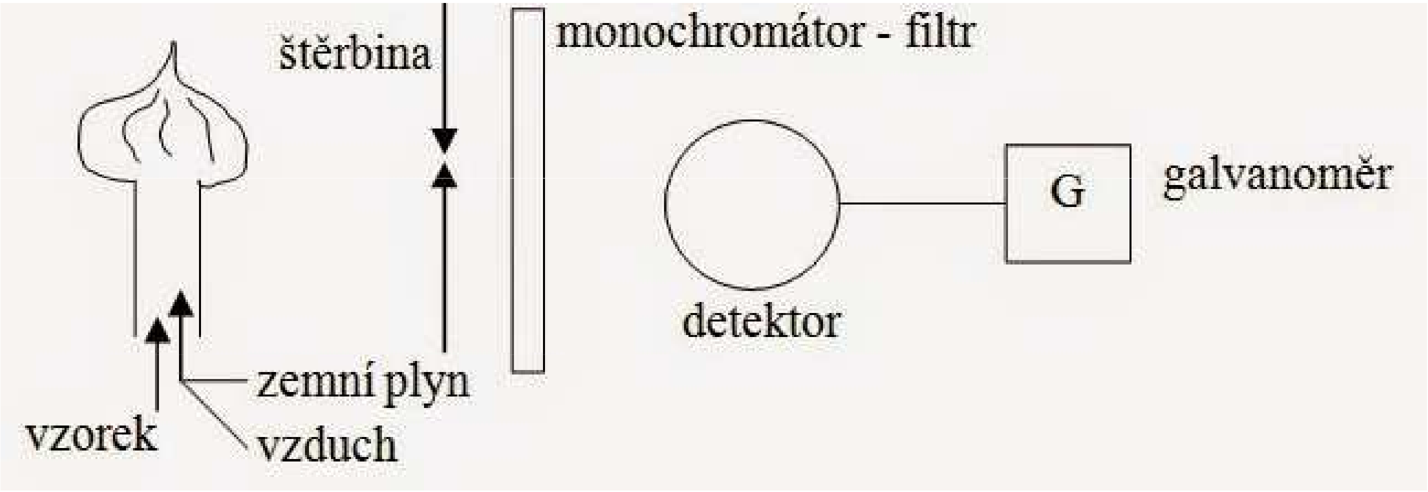


Plamenová fotometrie

Roztoková analýza

destruktivní

Stanovení snadno ionizovatelných prvků Na, Li, K, Mg a Ca



Atomová emisní spektrometrie s indukčně vázaným plazmatem (ICP-OES)

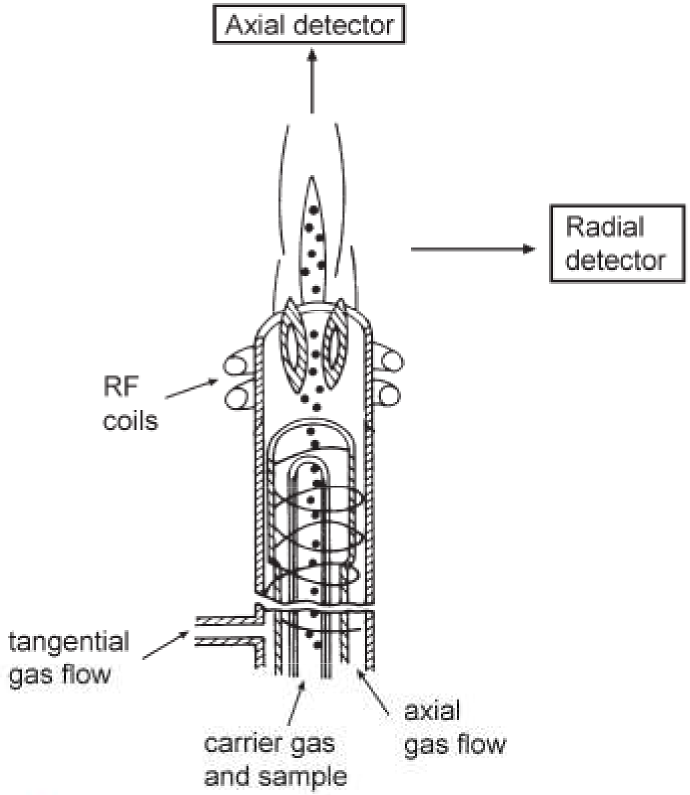


Figure 2.4 Schematic diagram of an ICP torch. The sample is carried into the torch by the carrier argon gas, and is ignited by radio-frequency heating from the RF coils. The tangential argon flow lifts the flame from the burner, preventing melting. The position of the detector in axial or radial mode is shown. (From Pollard *et al.*, 2007; Fig. 3–3, by permission of Cambridge University Press.)

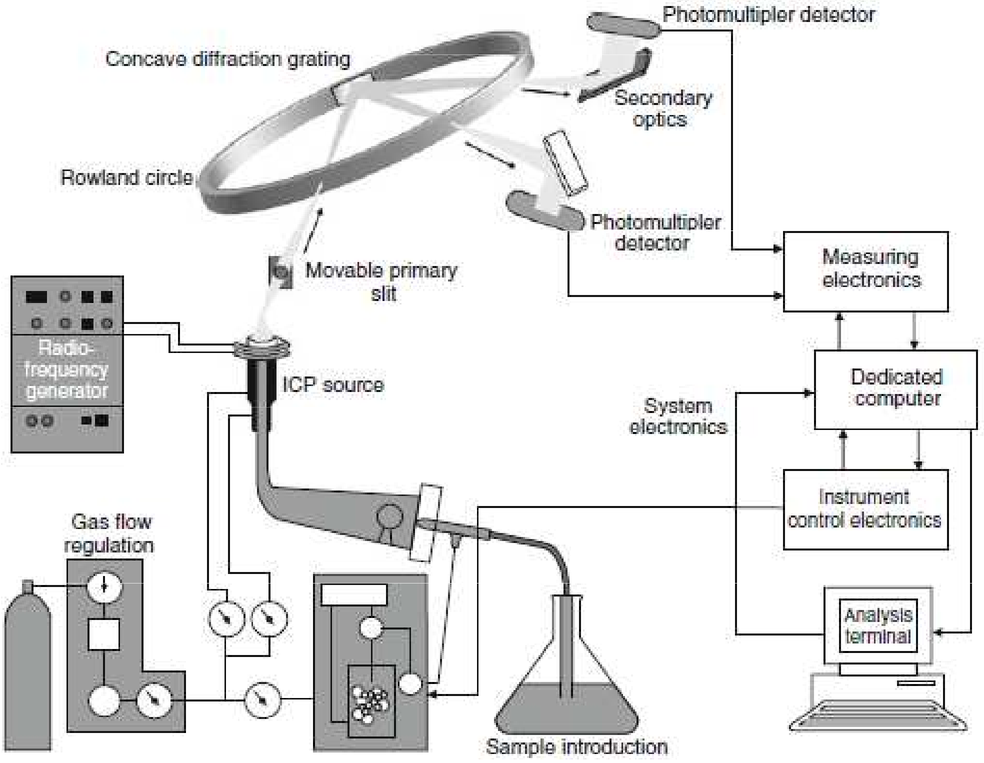
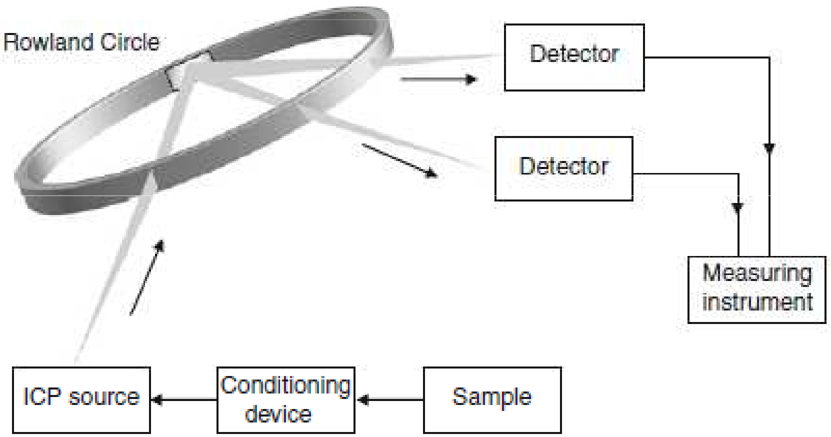


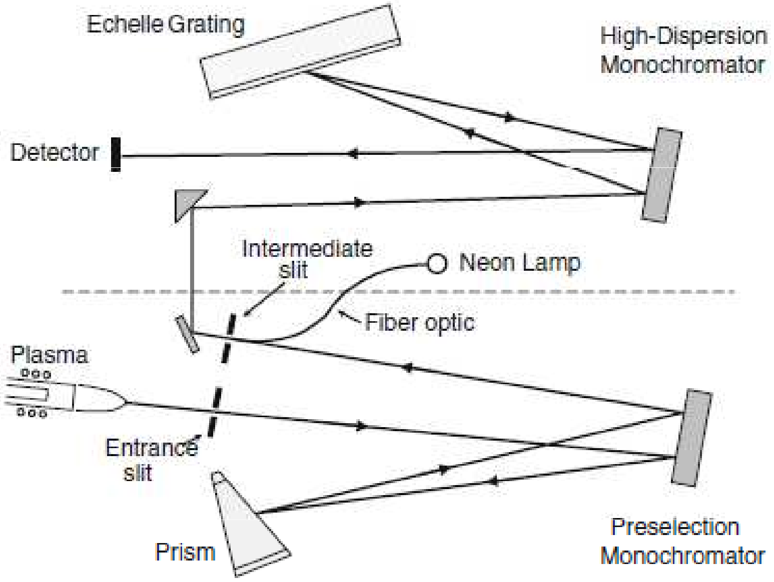
Fig. 34.2 Inductively coupled plasma polychromator (From SKOOG. Principles of Instrumental Analysis, 5E. © 1998 Brooks/Cole, a part of Cengage Learning, Inc. Reproduced by permission. www.cengage.com/permissions)

Roztoková analýza

Destruktivní



(a) Simultaneous Multichannel Detector



(b) Sequential Detector

Fig. 34.4 Simultaneous multichannel (a) and sequential detectors (b) ((a) Adapted from SKOOG. Principles of Instrumental Analysis, 5E. © 1998 Brooks/Cole, a part of Cengage Learning, Inc. Reproduced by permission. www.cengage.com/permissions (b) Image used with permission from PerkinElmer, Inc., Waltham, MA)

Multichannel Spectrometers

Spectrographs

Charge-Coupled Devices – 2 CCD's – one for vis and one for UV.

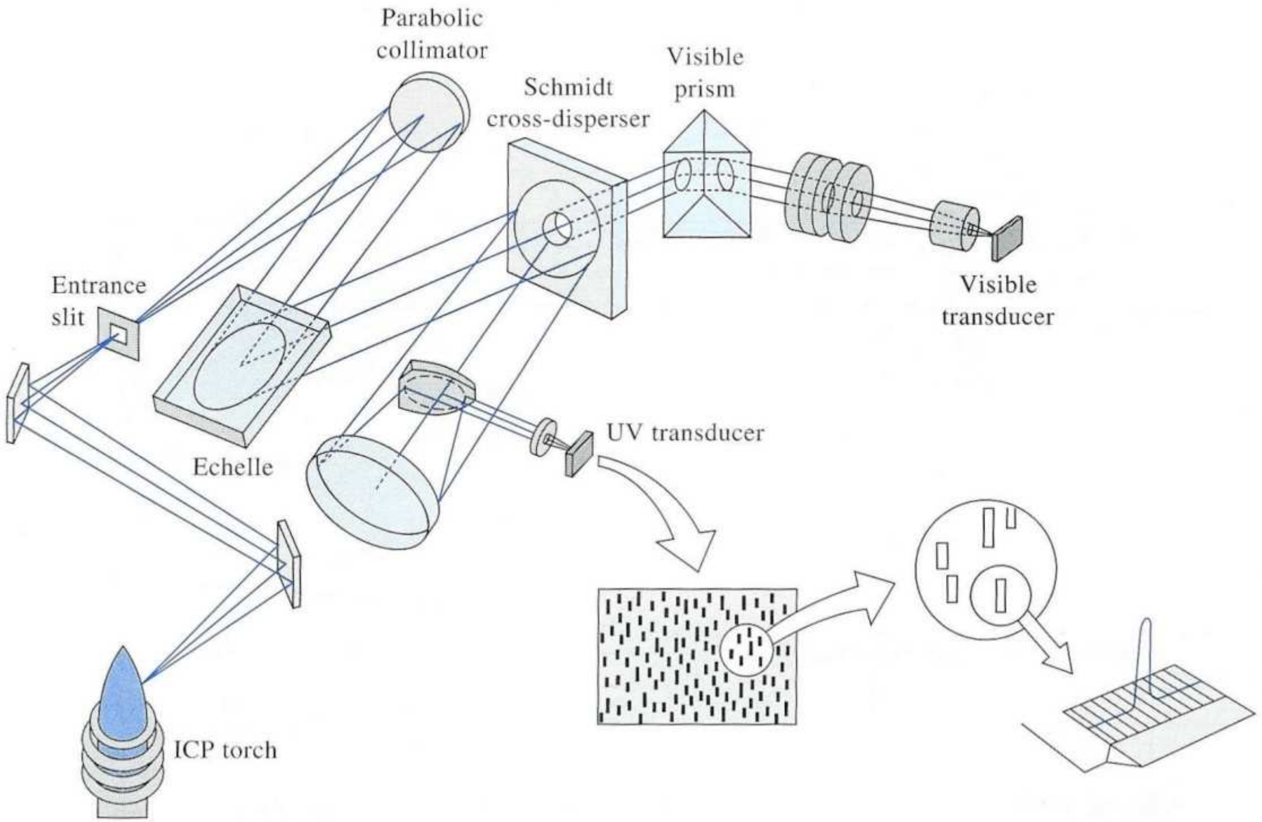
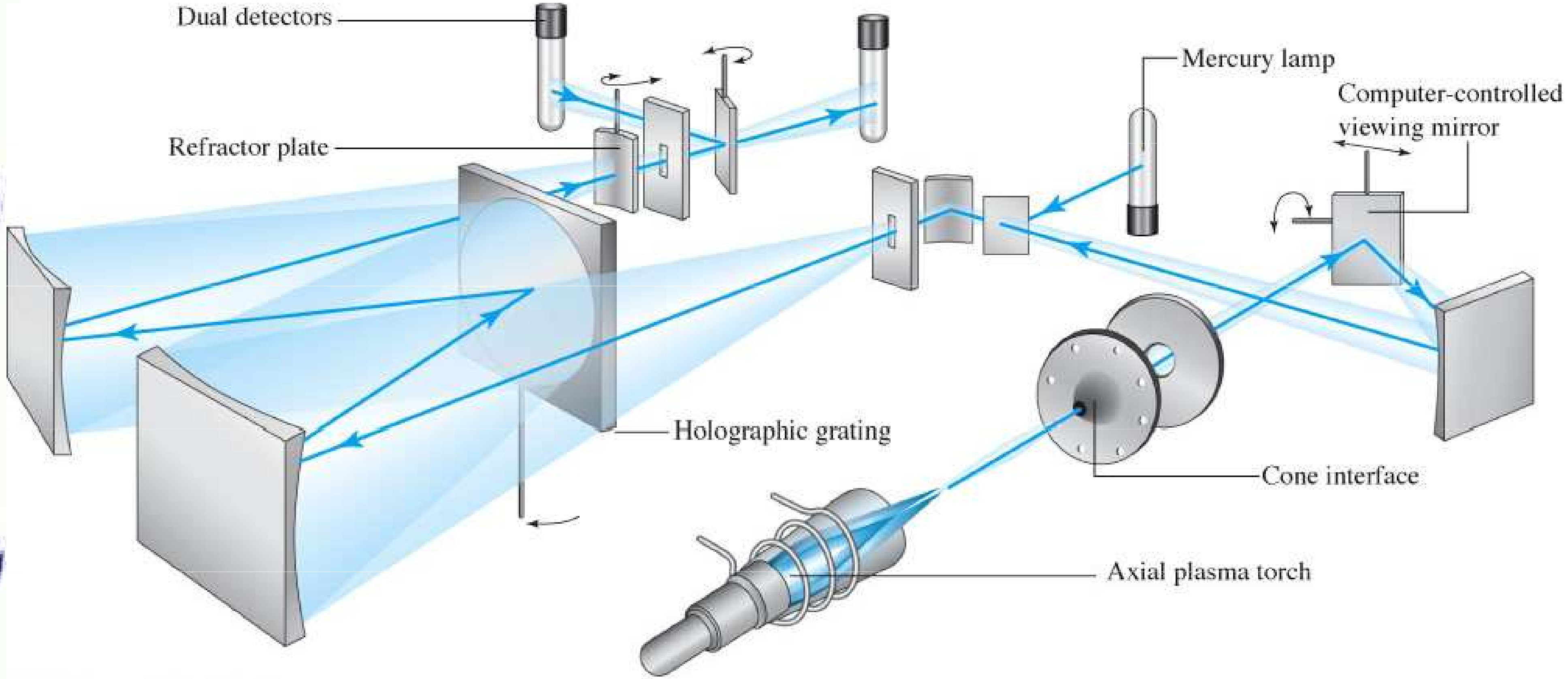


FIGURE 10-11 An echelle spectrometer with segmented array of CCDs. (From T. W. Barnard et al., *Anal. Chem.*, **1993**, *65*, 1231. Figure 1, p. 1232. Copyright 1993 American Chemical Society.)

Inductively Coupled Plasma Spectrometry

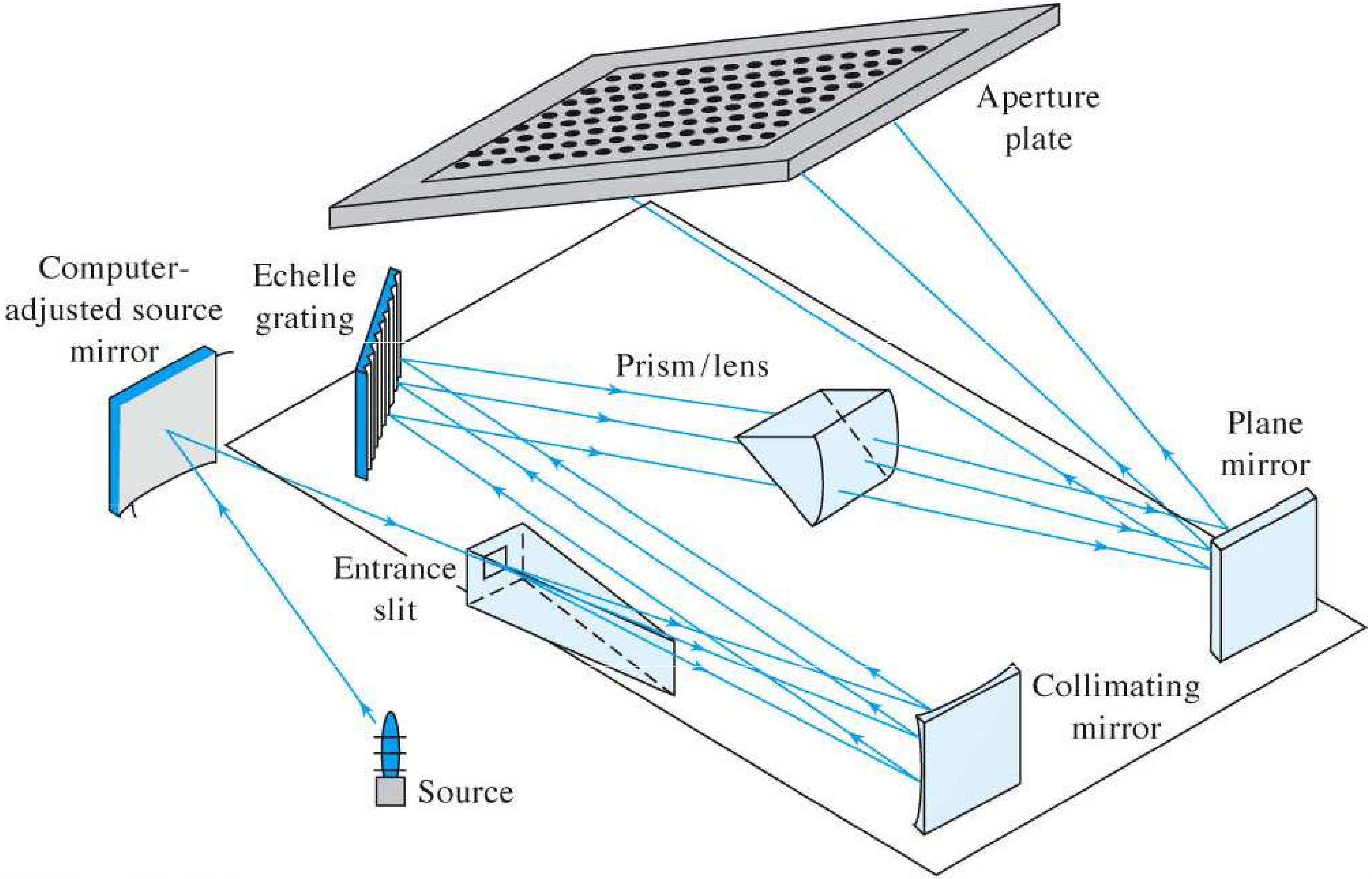
Sequential Instrument



Inductively Coupled Plasma Spectrometry

Sequential Instruments

Scanning Echelle Spectrometers



© 2007 Thomson Higher Education

Inductively Coupled Plasma Spectrometry

Multichannel Spectrometers

Polychromators

Advantages

-high precision

Disadvantages

-cost

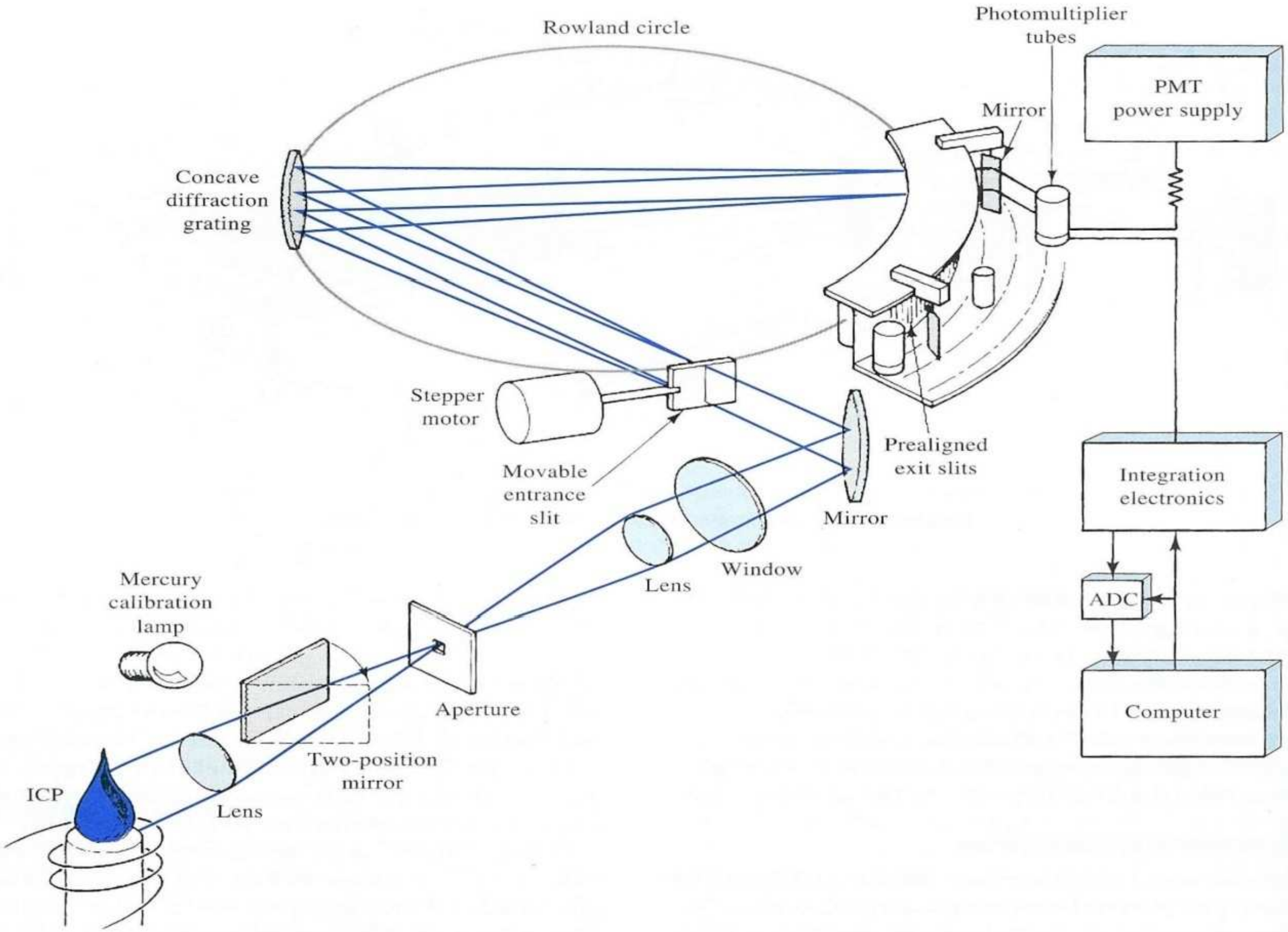


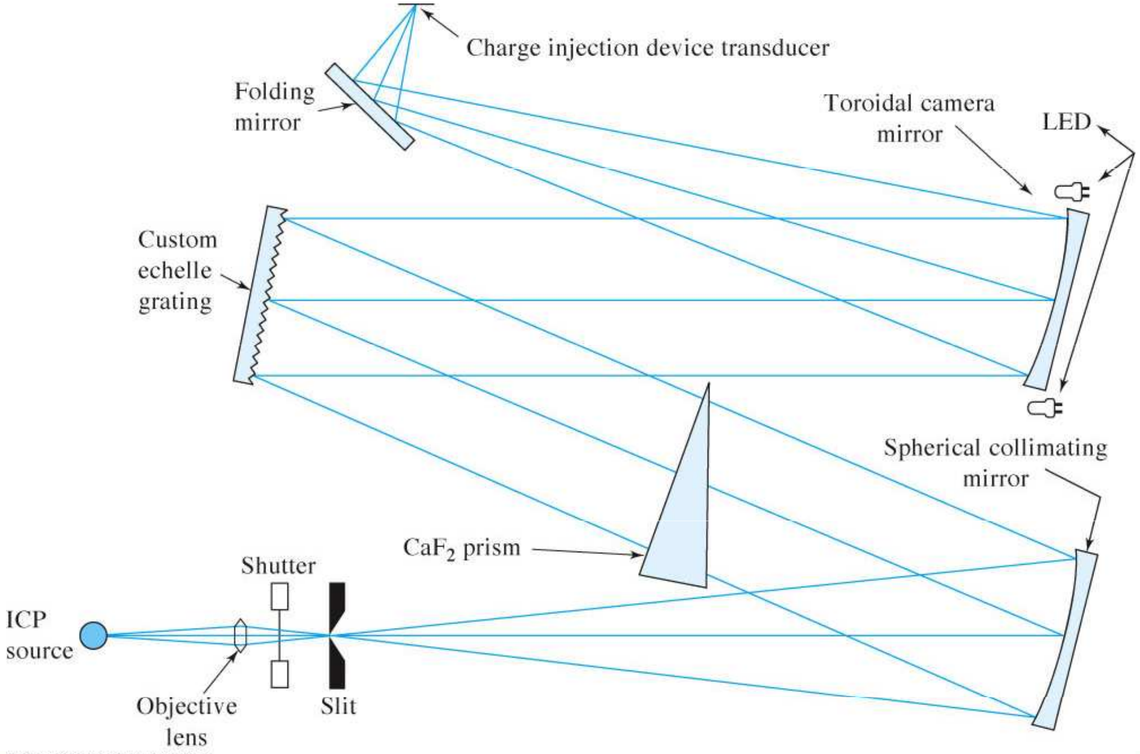
FIGURE 10-8 Direct-reading ICP emission spectrometer. The polychromator is of the Paschen-Runge design. It features a concave grating and produces a spectrum around a Rowland circle. Separate exit slits isolate each spectral line, and a separate photomultiplier tube converts the optical information from each channel into an electrical signal. Notice the radial viewing geometry. PMT = photomultiplier tube. (From J. D. Ingle Jr. and S. R. Crouch, *Spectrochemical Analysis*, p. 241, Upper Saddle River, NJ: Prentice-Hall, 1988, with permission.)

Inductively Coupled Plasma Spectrometry

Multichannel Spectrometers

Spectrographs

Charge-Injection Devices – based on echelle spectrometers and 2D array devices



Inductively Coupled Plasma Spectrometry

Multichannel Spectrometers

Spectrographs

Charge-Coupled Devices – 2 CCD's – one for vis and one for UV.

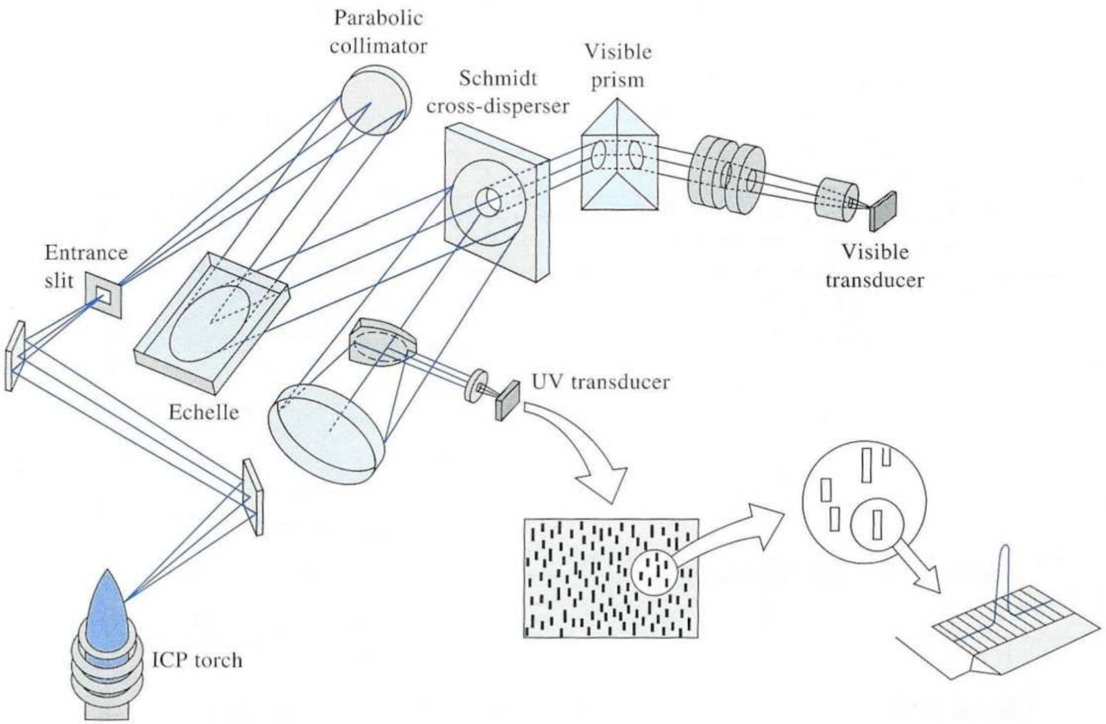
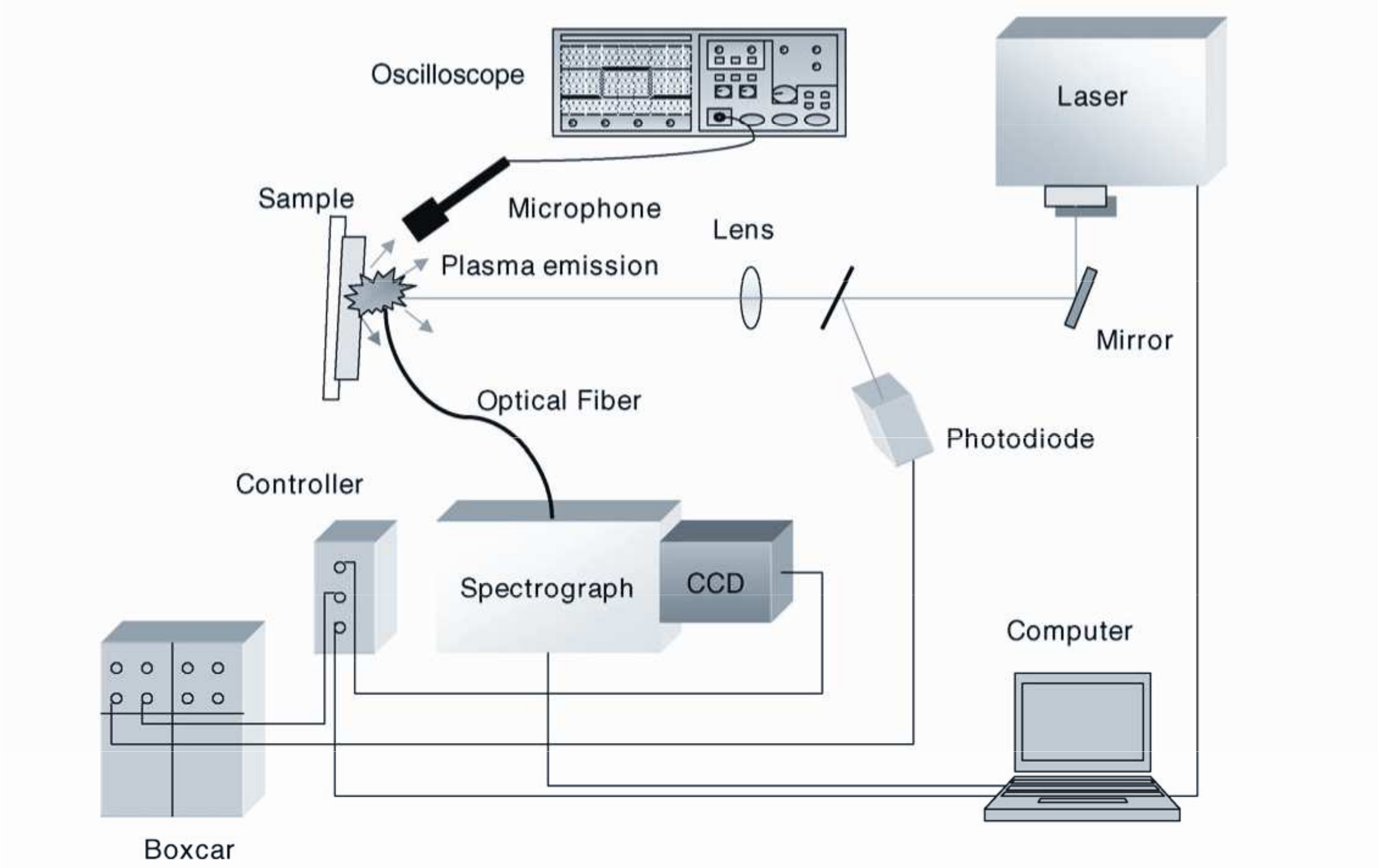


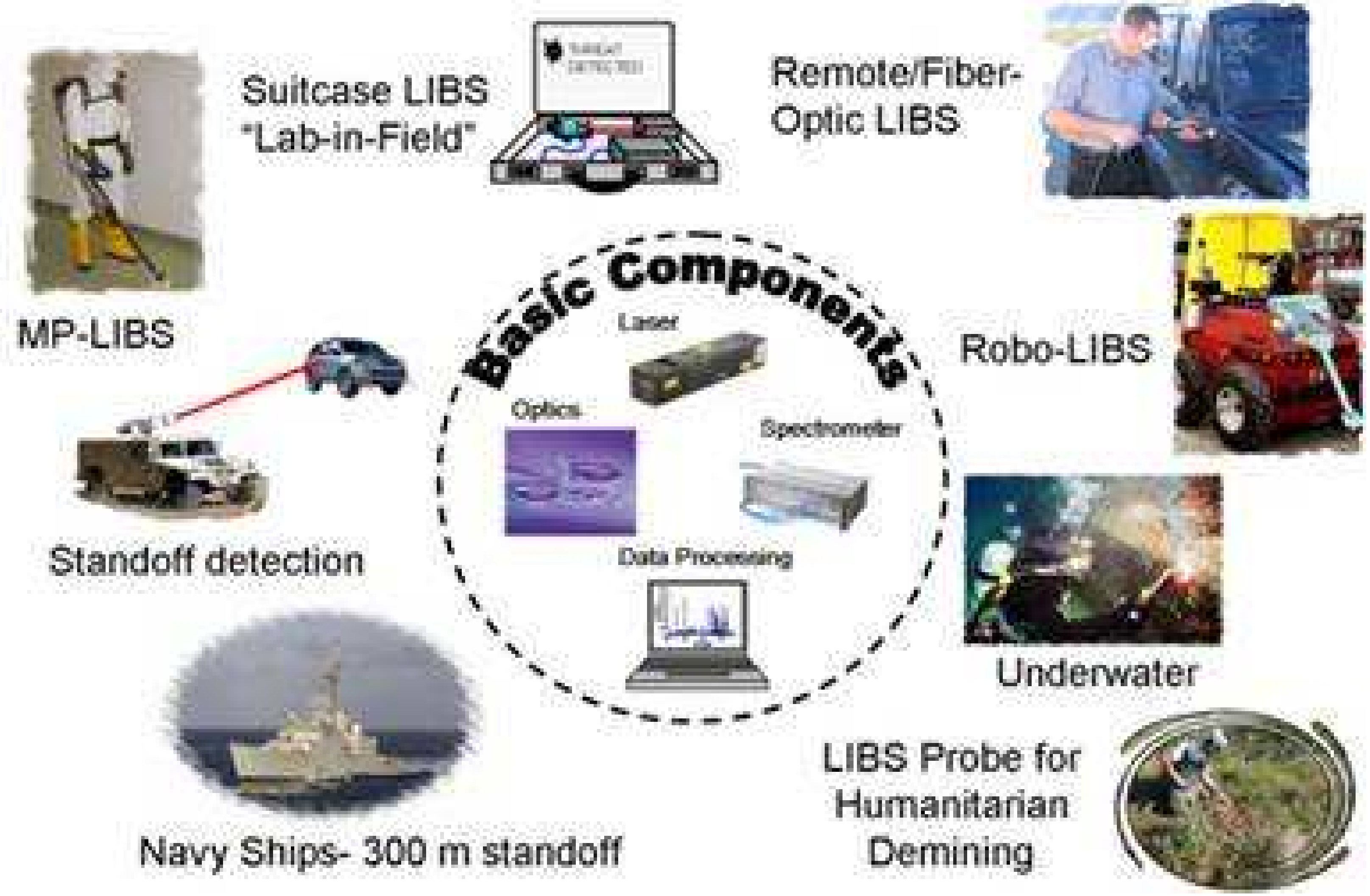
FIGURE 10-11 An echelle spectrometer with segmented array of CCDs. (From T. W. Barnard et al., *Anal. Chem.*, **1993**, 65, 1231. Figure 1, p. 1232. Copyright 1993 American Chemical Society.)

Laser Induced Breakdown Spectroscopy

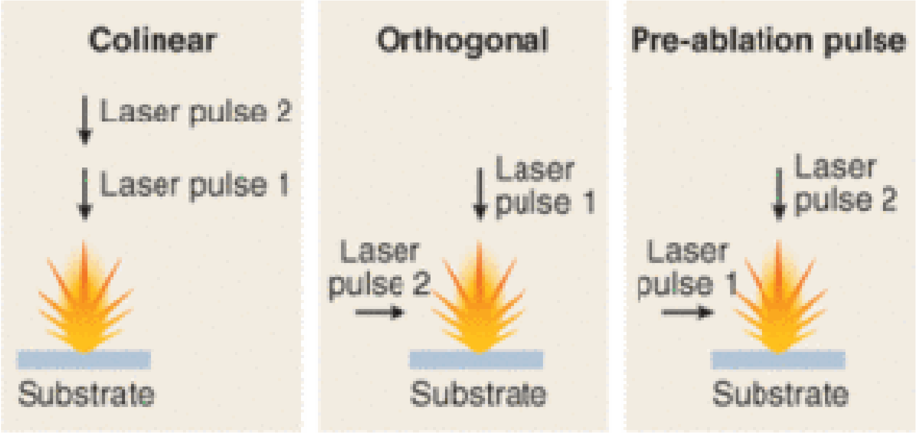


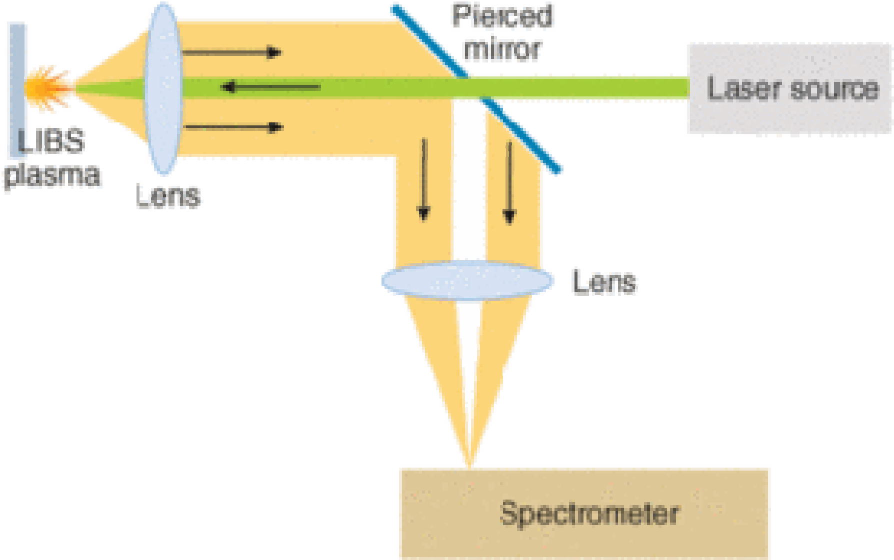
Schematic diagram of the experimental set-up used for the LIBS experiments and for the photoacoustic measurements.

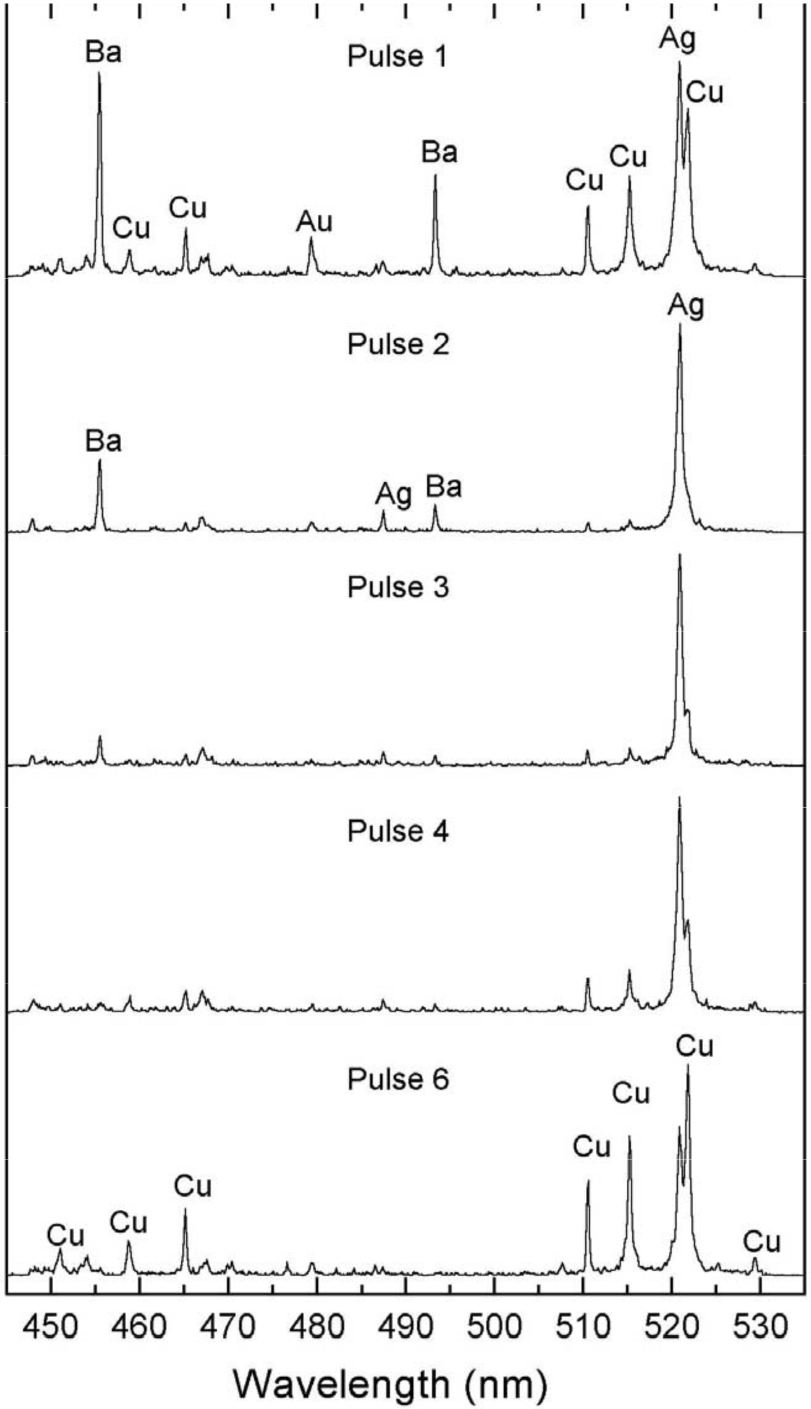
LIBS


Suitcase LIBS
"Lab-in-Field"
Remote/Fiber-Optic LIBS
MP-LIBS
Basic Components
Laser
Optics
Spectrometer
Data Processing
Robo-LIBS
Standoff detection
Underwater
Navy Ships- 300 m standoff
LIBS Probe for Humanitarian Demining

LIBS

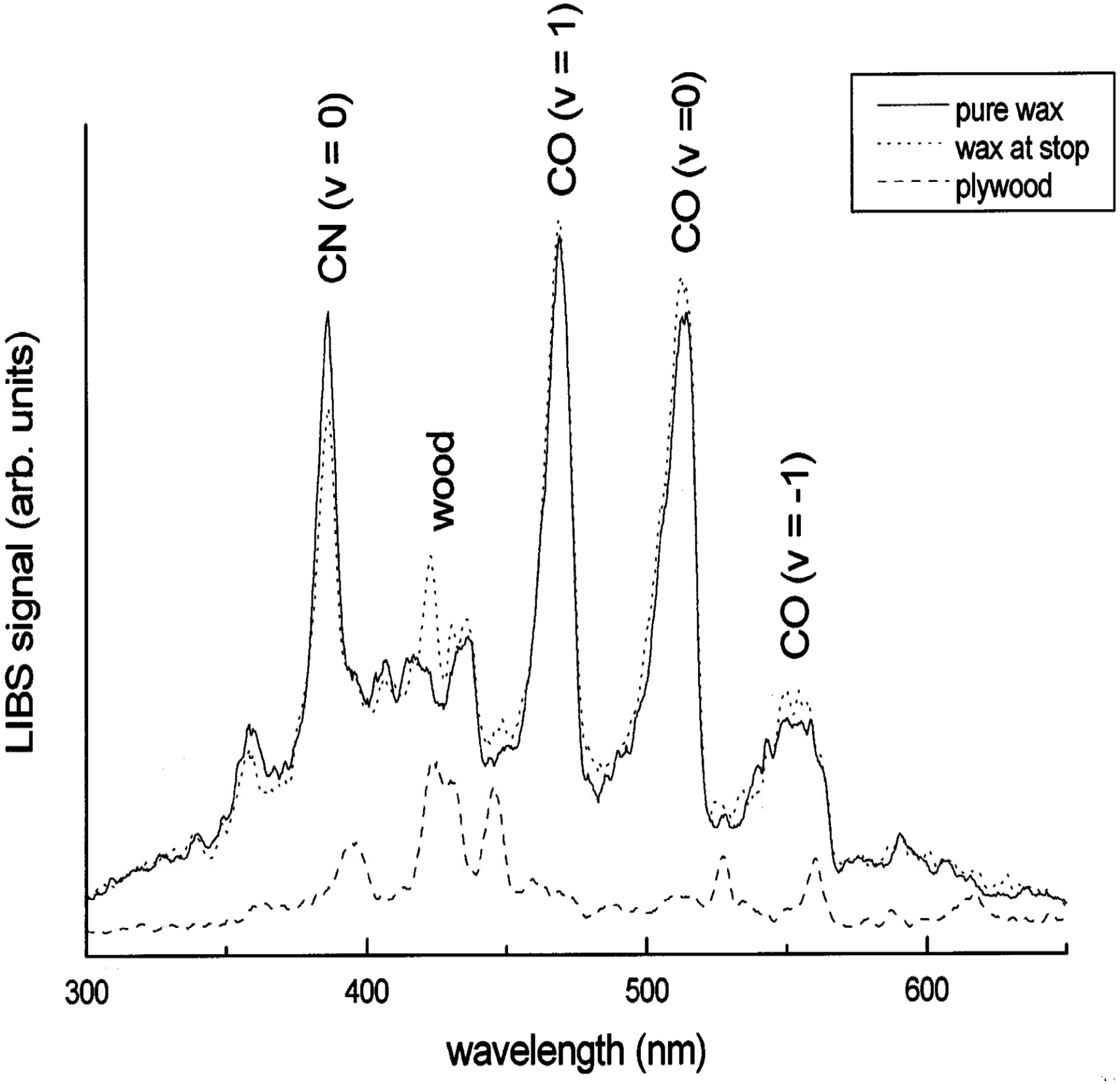






Daguerrotypie, 19. stol.

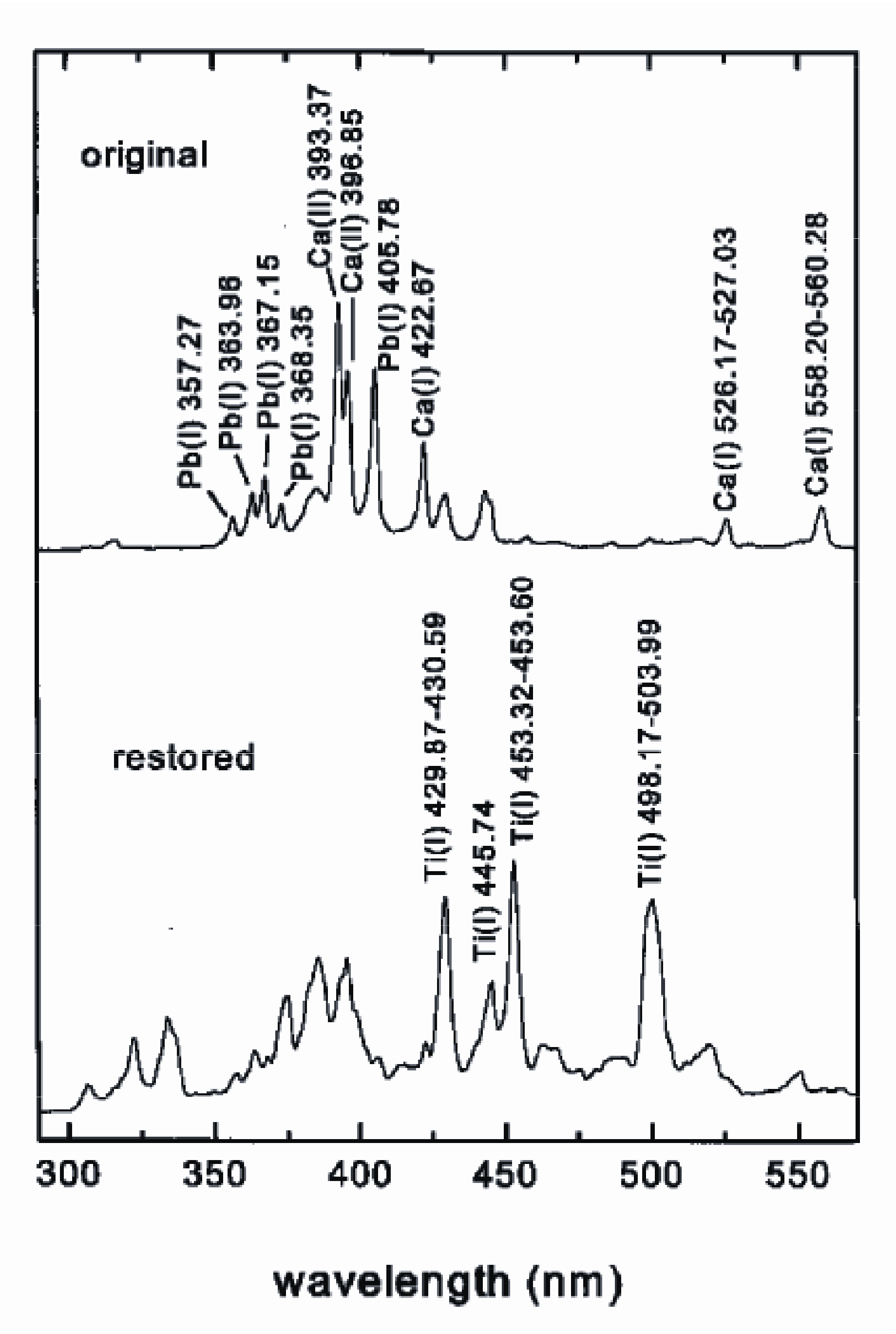
Monitoring odstraňování starých nátěrů



Odstraňování vosku z pláten nebo dřeva (překližka) je časově náročné. Proces lze snadno automatizovat použitím laseru a detekce LIBS.

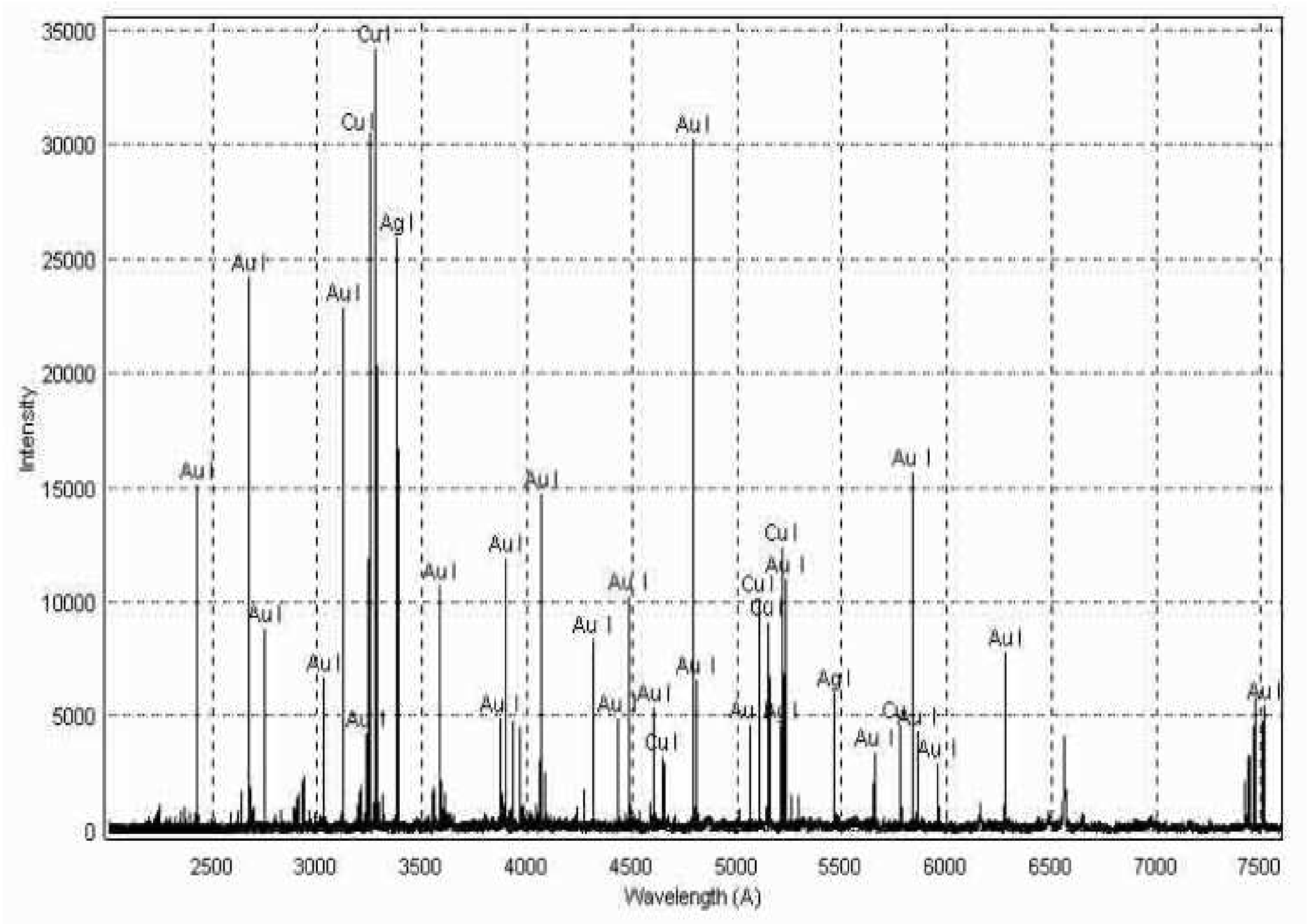
LIBS spektra vosku vykazují zřetelné pásy CO a CN. Po dosažení spodní vrstvy se objevuje pík 423 nm a čištění je ukončeno.

Sledování restaurátorských zásahů



LIBS spektra originální malby a restaurovaných částí olejomalby.

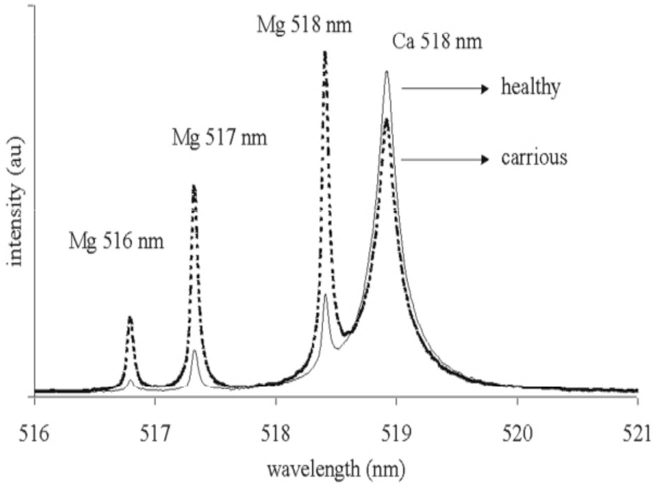
Kovy a slitiny

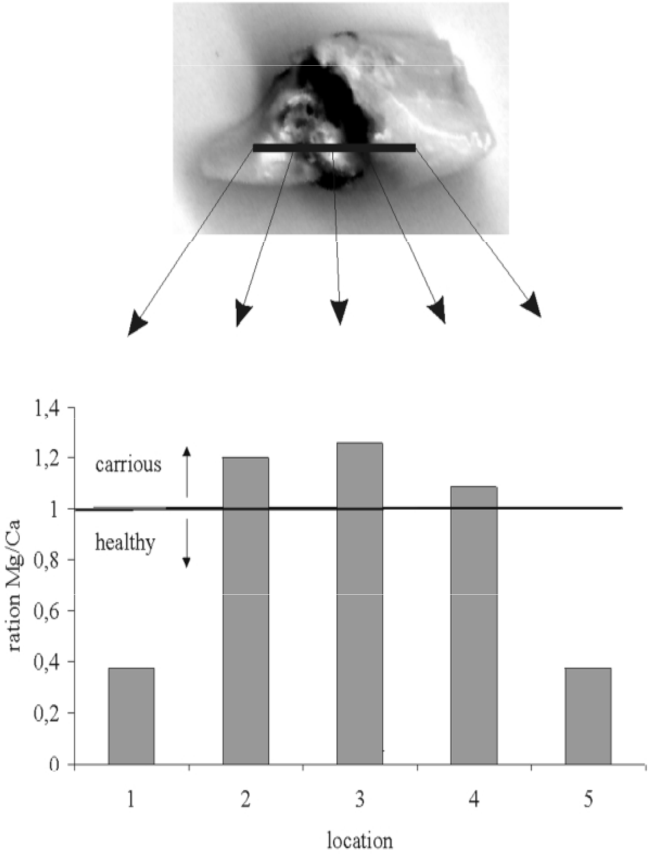


LIBS analýza slitiny Au.

Kosti a zuby

Obsah Mg a Ca v zubní tkáni poškozené kazem. Zvýšená koncentrace Mg zřetelně indikuje postižené partie.





Autenticita výrobků z korálu

korál vs. vápenec

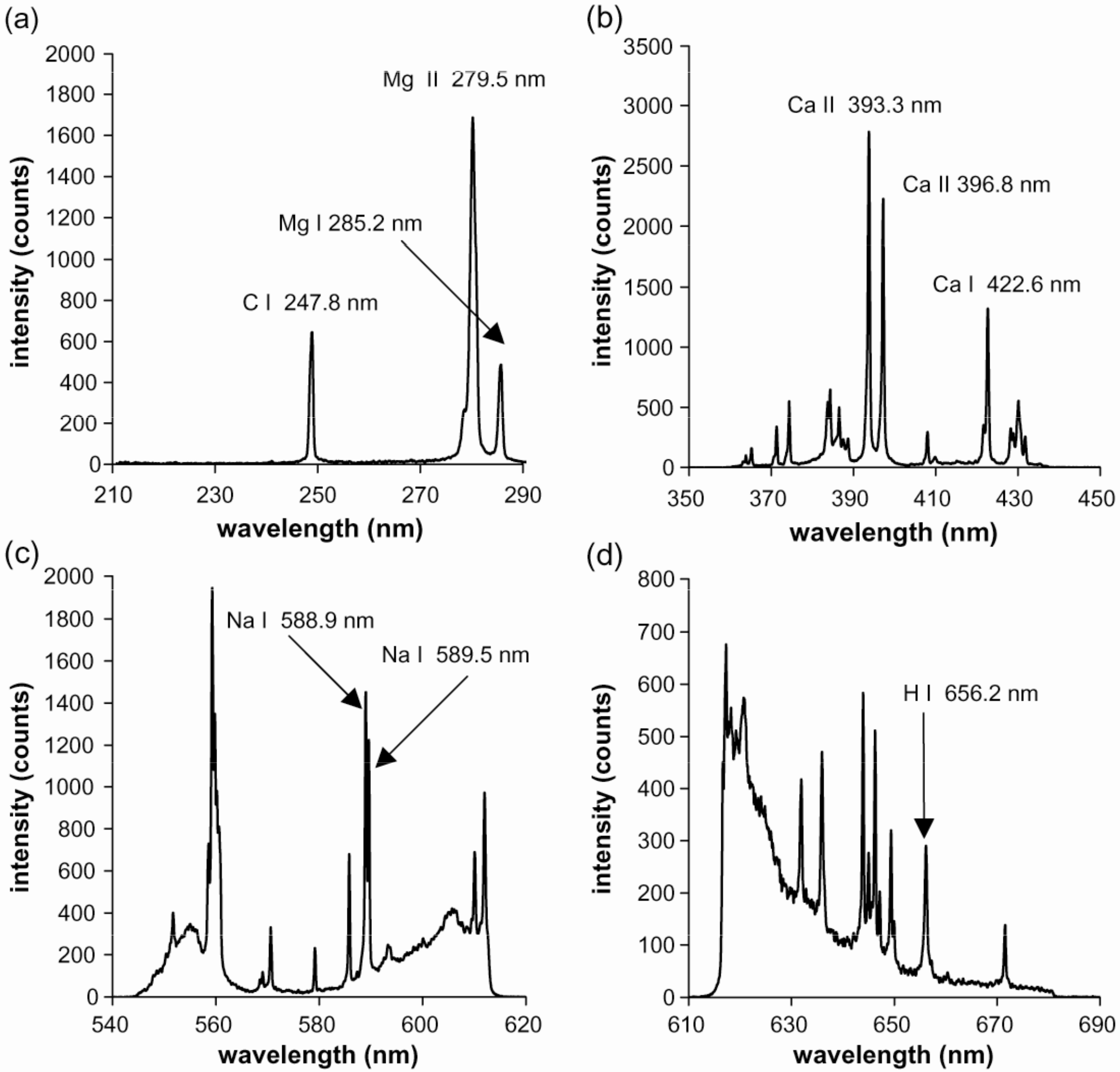
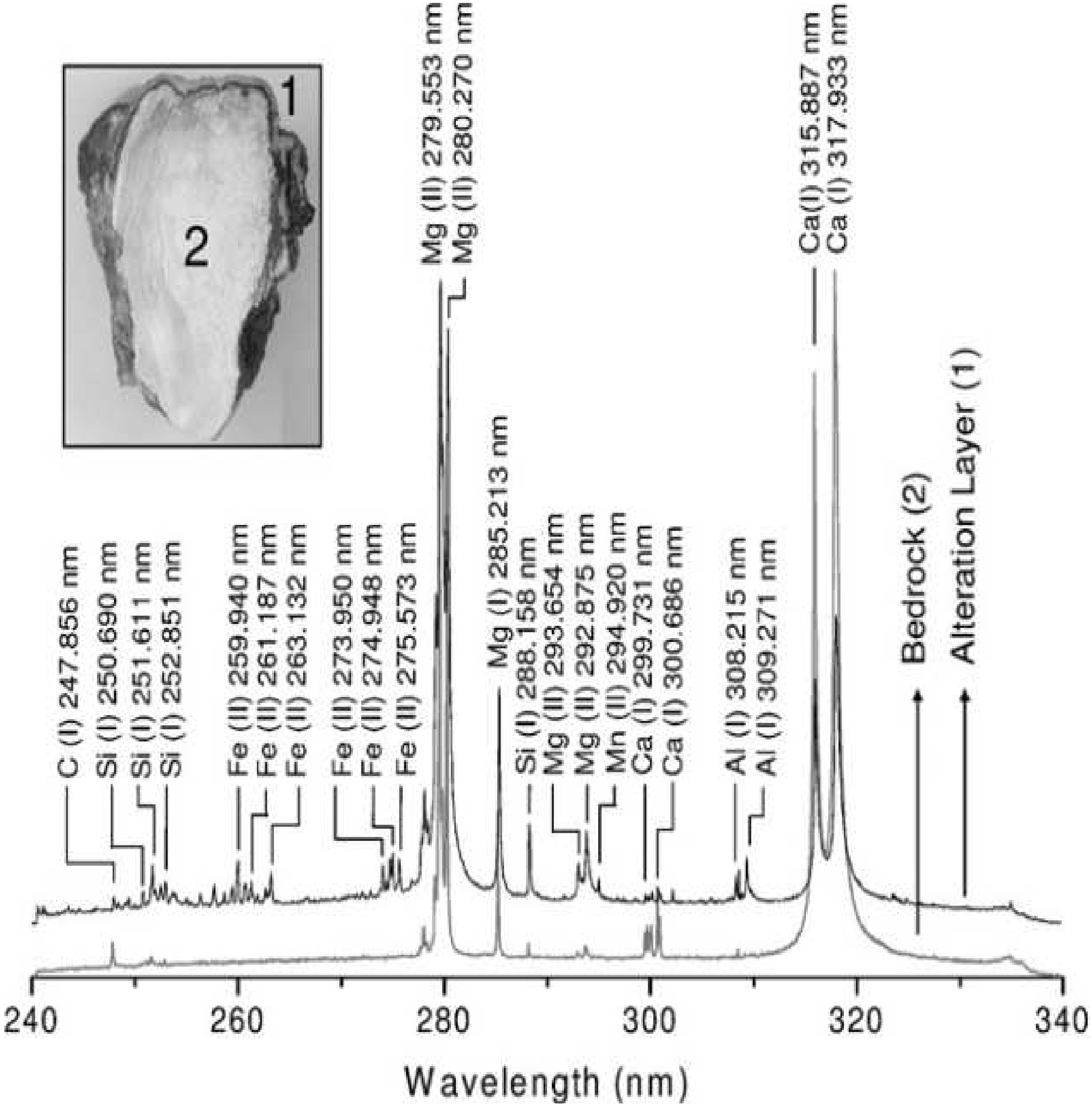


Fig. 5. Emission spectra of a fresh white coral sample in a low pressure plasma of 1.3 kPa in the wavelength region between (a) 220 and 290 nm, (b) 370 and 440 nm, (c) 550 and 620 nm and (d) 620 and 680 nm.

Čištění kamene



LIBS řezu krápníkem (znečištěný povrch)

LIBS

Molekulové pásy →

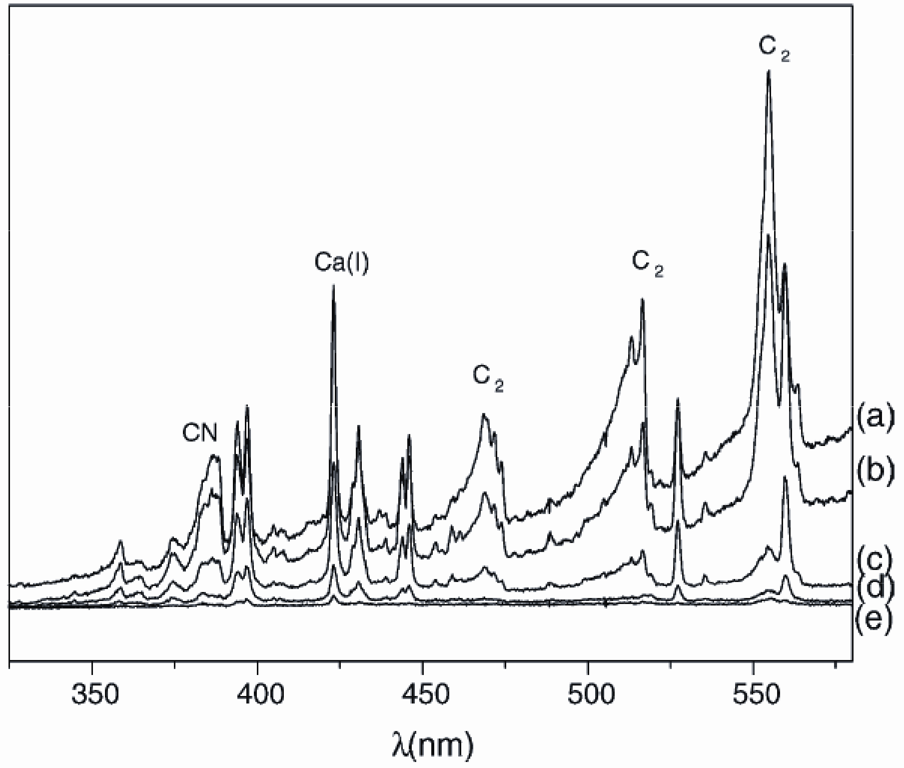
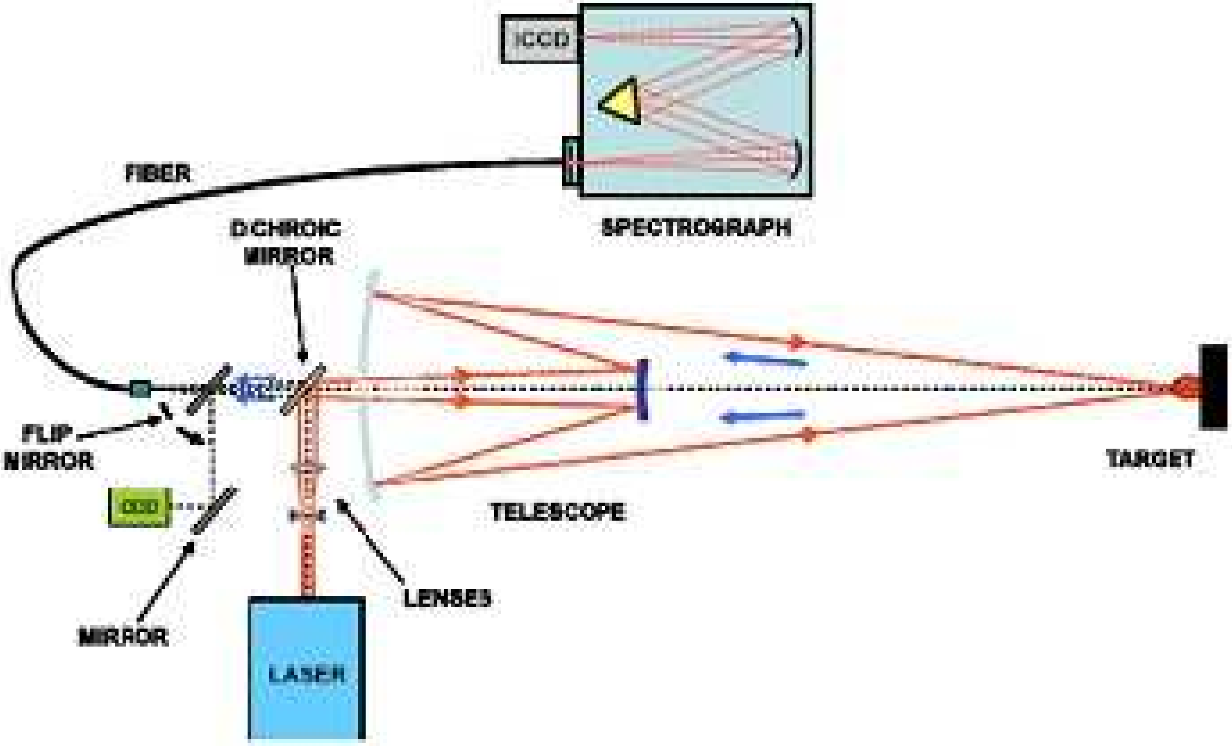


Fig. 6. Evolution of LIBS spectrum with the number of pulses during the removal of black paint on wood: (a) pulse 1, (b) pulse 2, (c) pulse 4, (d) pulse 7 and (e) pulse 9. Irradiation wavelength: 308 nm.

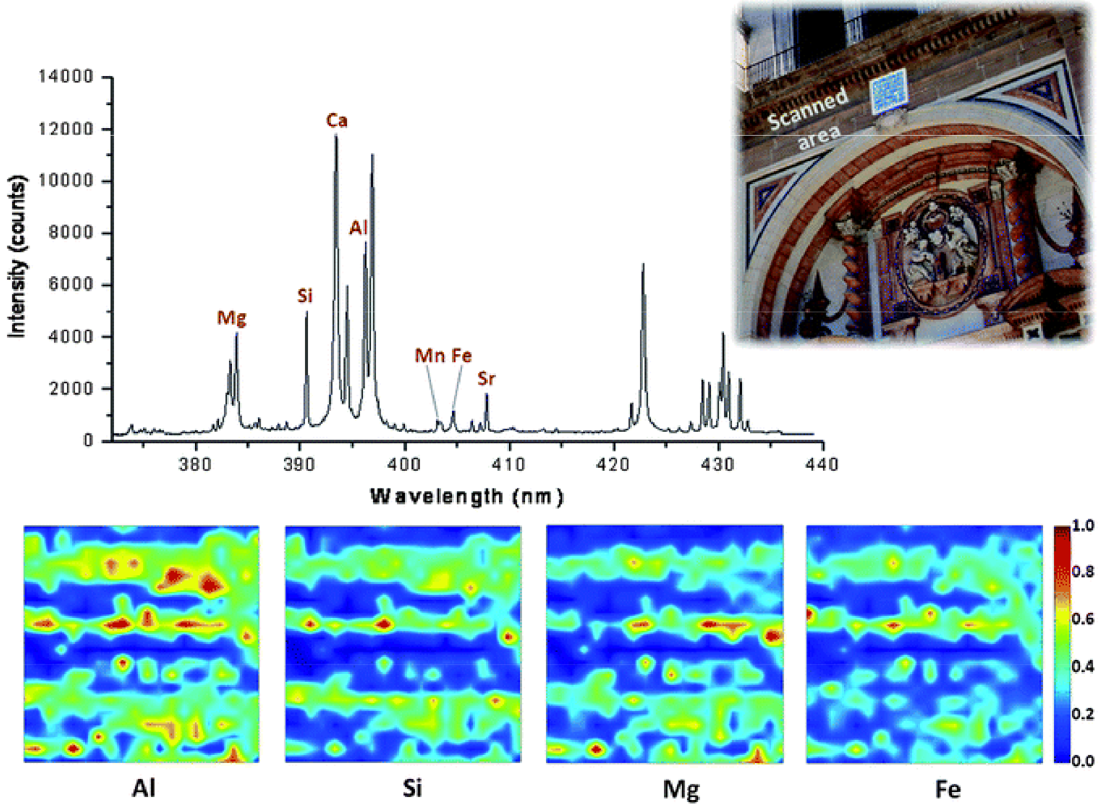
Spojení s Ramanovou spektrometrií

Stand-off LIBS

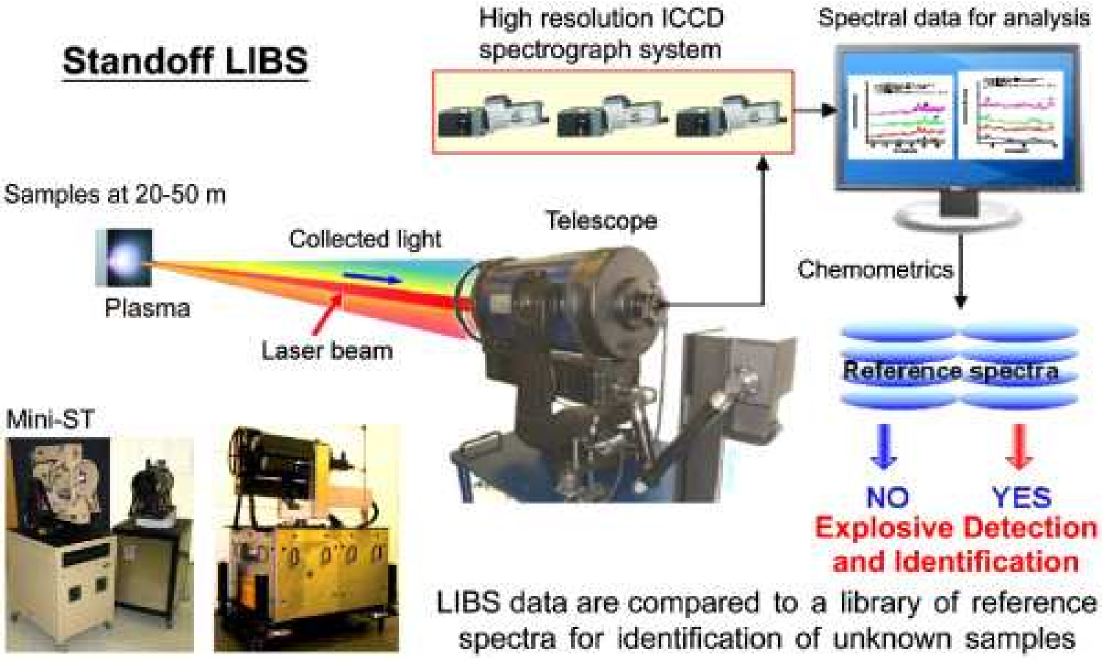


Malaga

Analýza portálu katedrály.

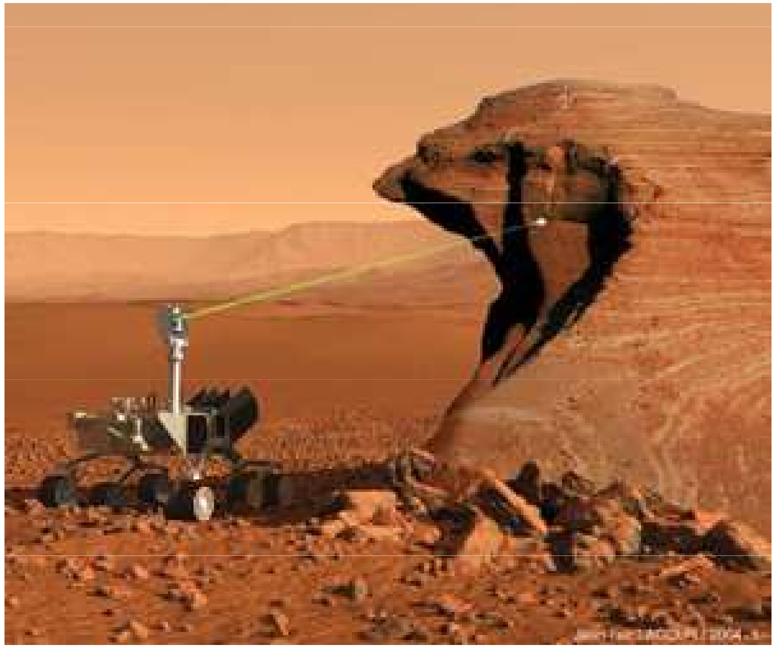


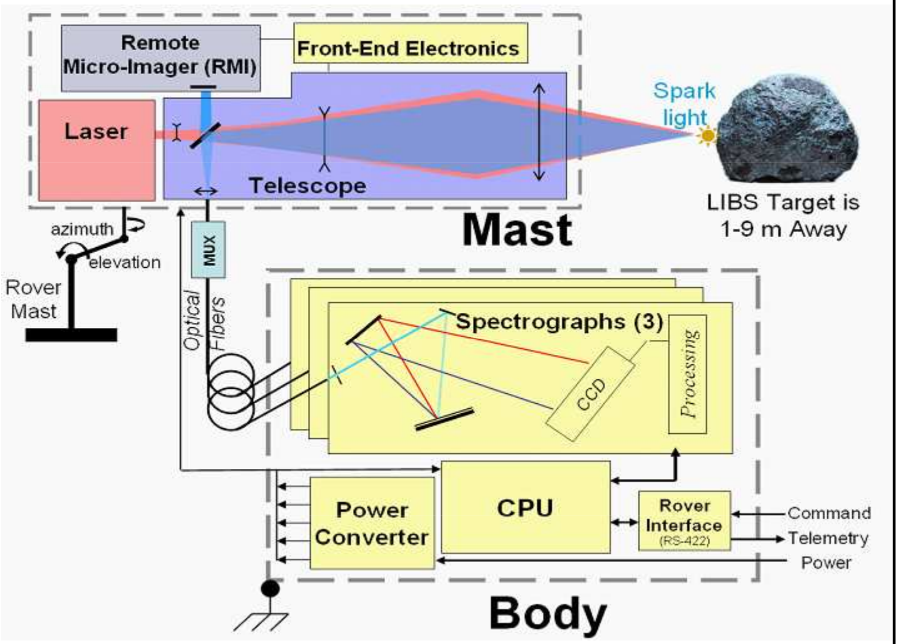
Stand-off LIBS



LIBS data are compared to a library of reference spectra for identification of unknown samples

ChemCam

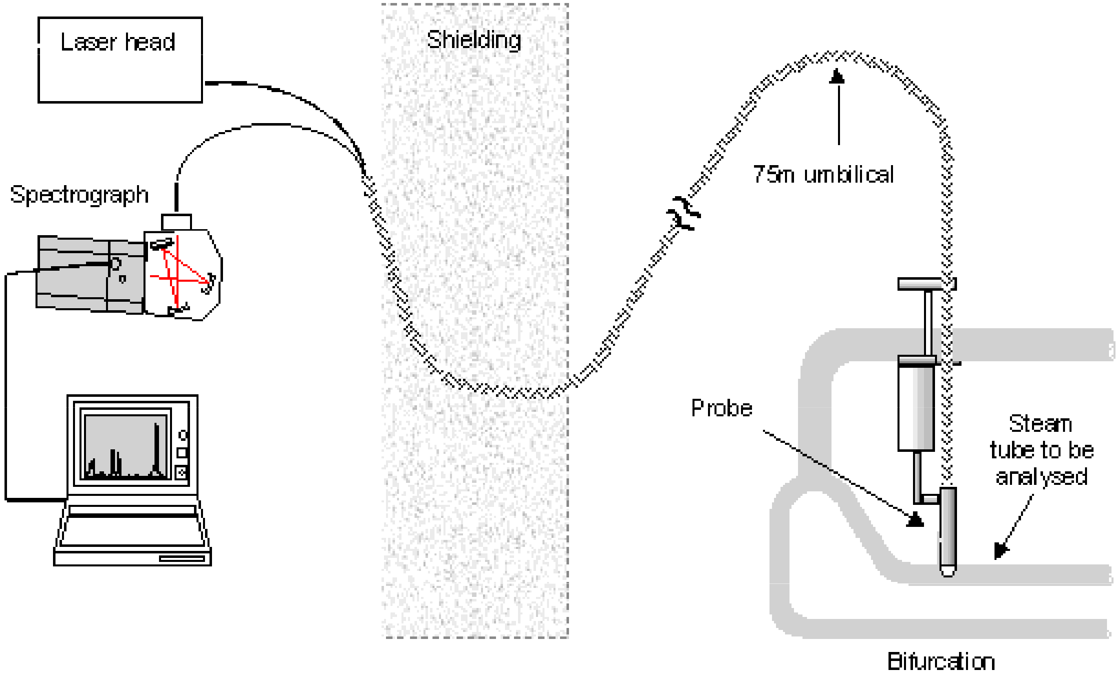




Spektrometrie laserem indukovaného plazmatu (LIBS)

Terénní a mobilní zařízení

remote LIBS



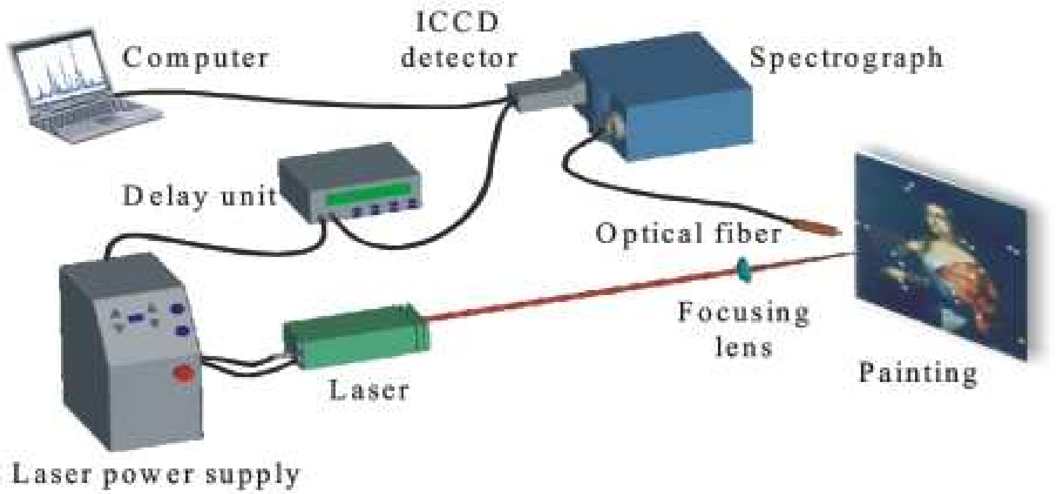
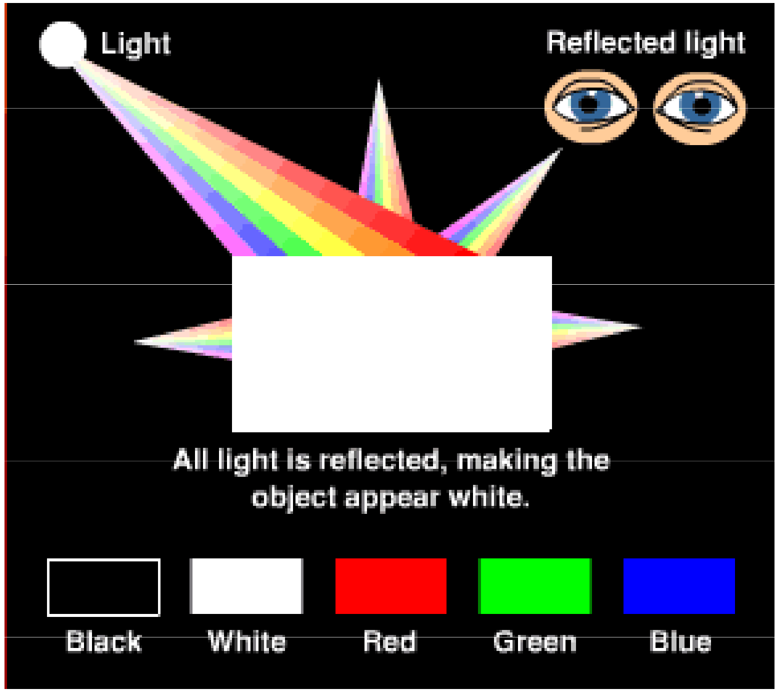
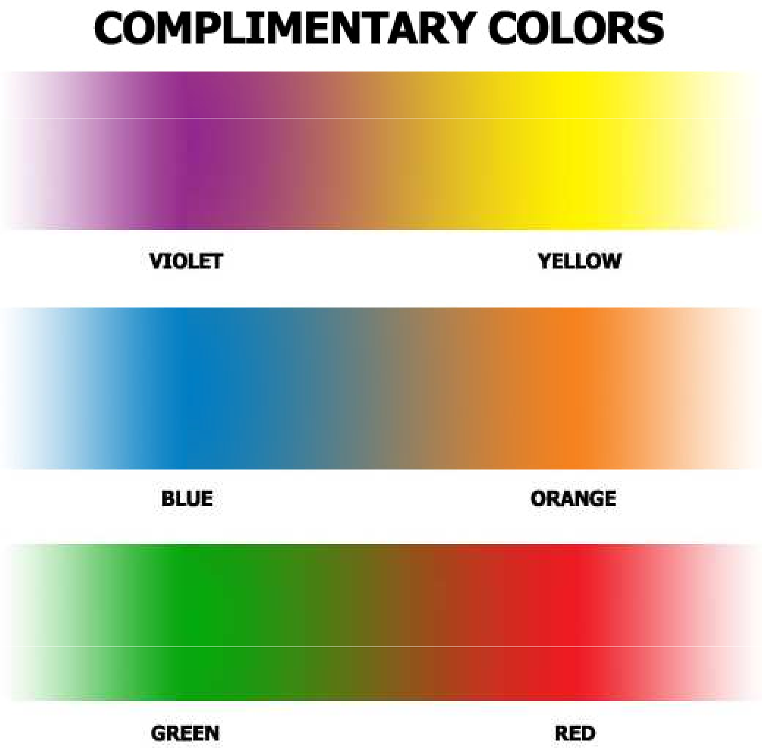


Fig.4.2.2: Typical experimental set-up for LIBS analysis.

© *Giakoumaki et al., in: "Handbook on the Use of Lasers in Conservation and Conservation Science", 2008.*

Absorpce záření





Absorpční spektrum

Soubor temných čar (pásů ve spojitém spektru světla), které vznikají při pohlcování záření látkou.

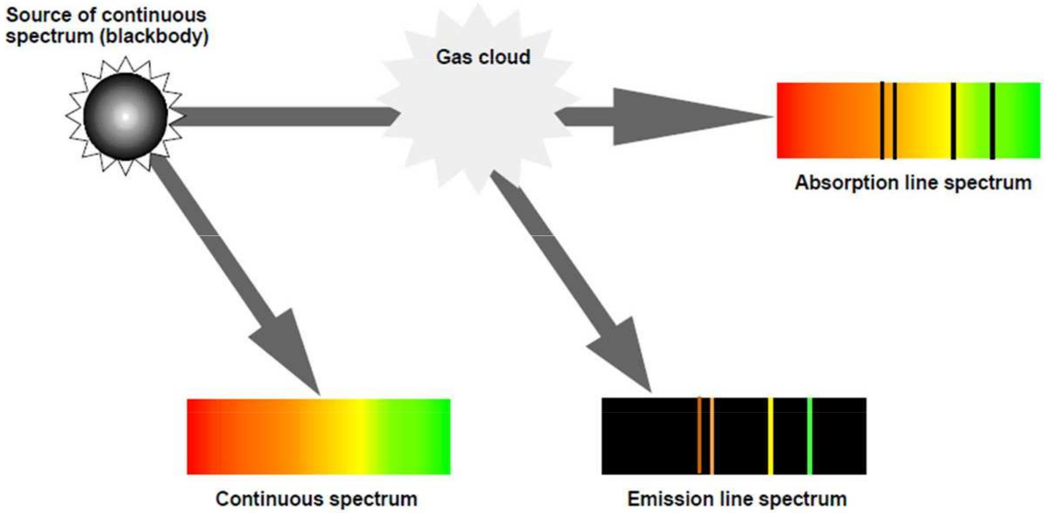
a) Čárové spektrum

b) Pásové spektrum

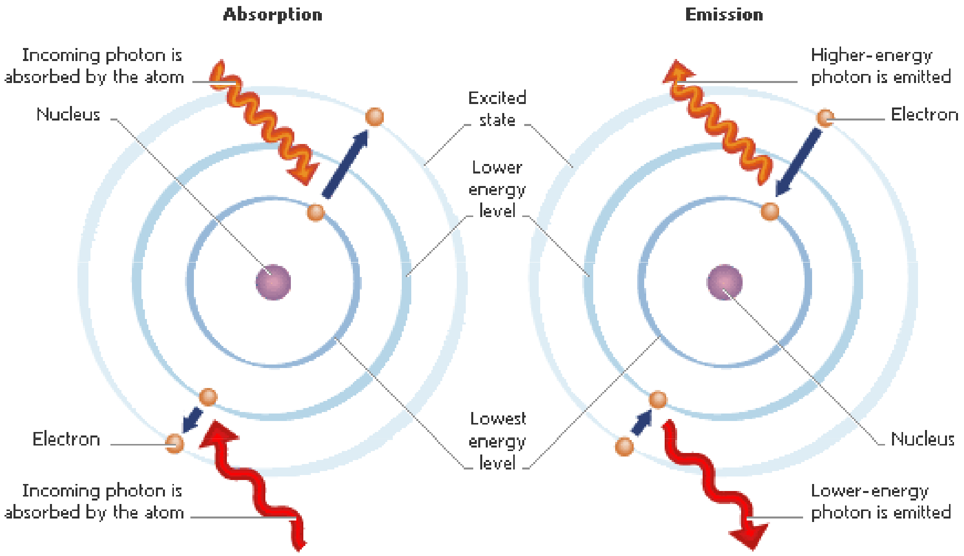
Na rozdíl od emisních spekter nemusíme vzorek látky rozžhavit na velmi vysokou teplotu.

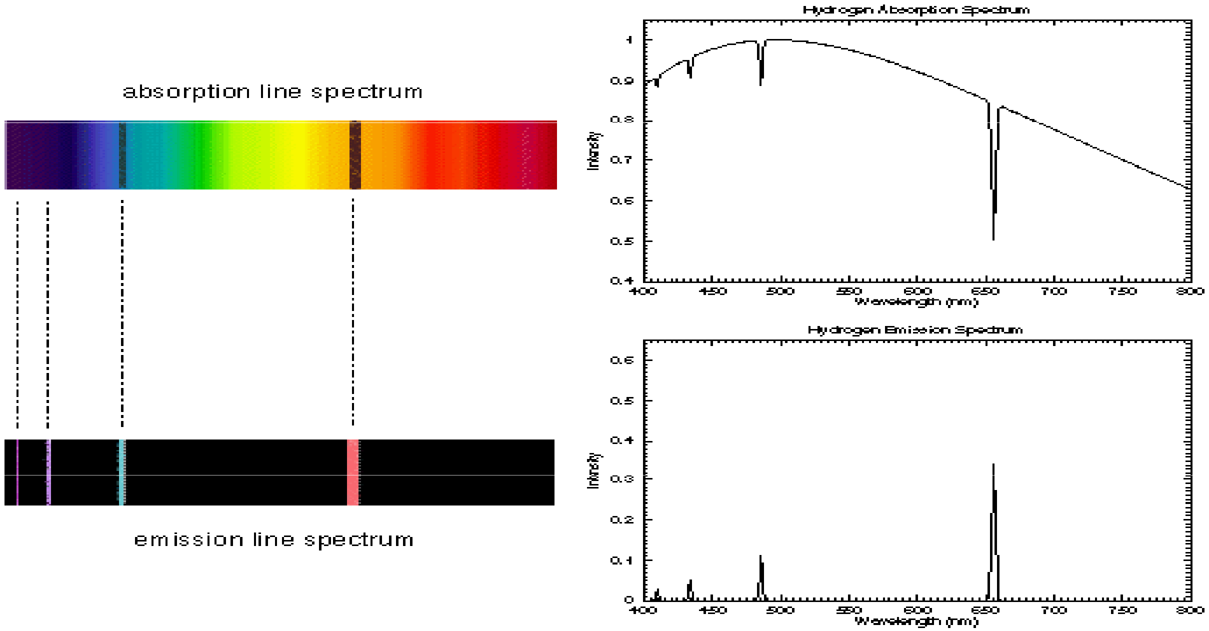
Kirchhoffův zákon

Těleso, které vysílá záření určitých vlnových délek, rovněž tyto vlnové délky absorbuje.
Sloučíme-li emisní a absorpční spektrum stejné látky, získáme spektrum spojité.



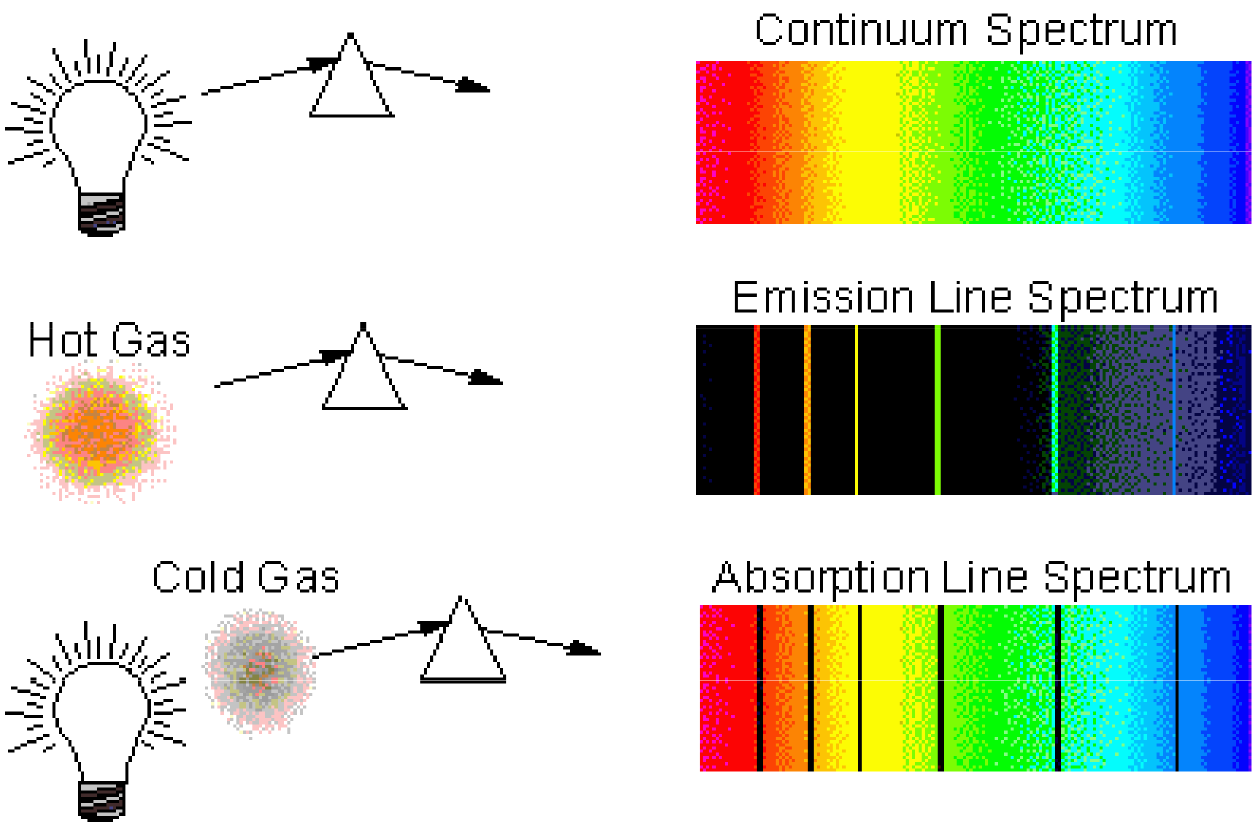
Zdroj záření, spektrum (čárové, pásové, spojité)



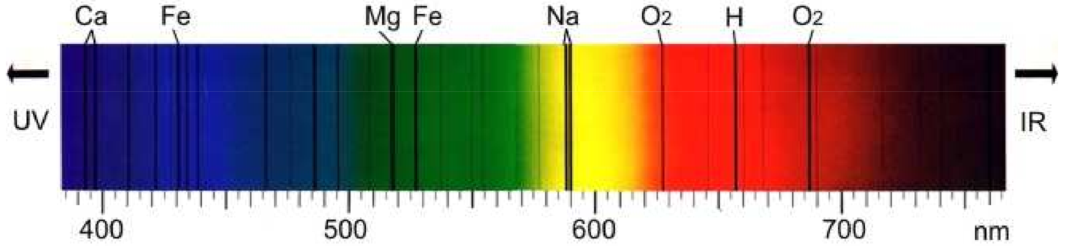


Two ways of showing the same spectra: on the **left** are pictures of the dispersed light and on the **right** are plots of the intensity vs. wavelength. Notice that the pattern of spectral lines in the absorption and emission line spectra are the **same** since the gas is the same.

Emisní a absorpční spektra



Fraunhoferovy čáry ve slunečním spektru jsou důsledkem absorpce světla atomy různých prvků ve sluneční atmosféře.



Emisní spektrum

Soubor frekvencí elektromagnetického záření vyzařovaného látkou.

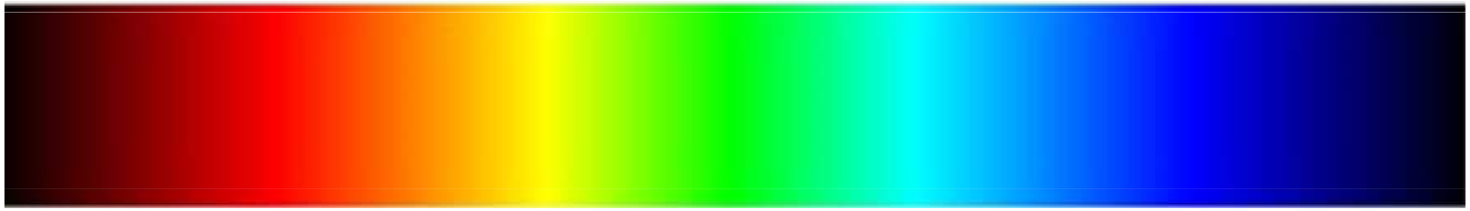
a) Spojité spektrum

b) Čárové spektrum

c) Pásové spektrum

Spojité spektrum

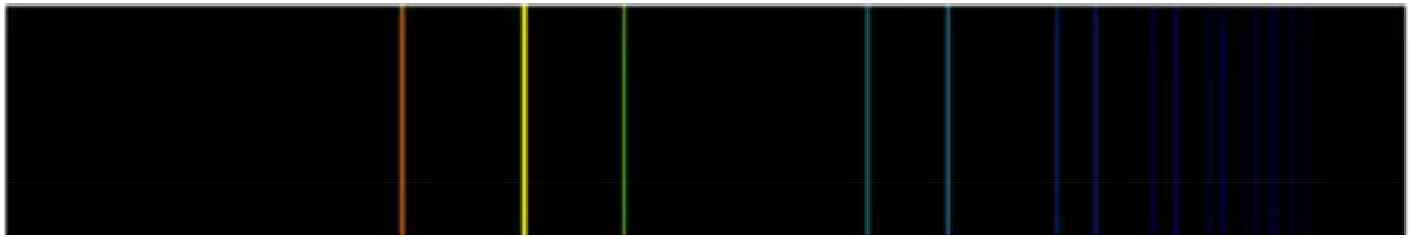
obsahuje elektromagnetické vlny všech vlnových délek v určitém intervalu, zdroj: rozžhavené pevné a kapalné látky(např. vlákno žárovky, roztavené kovy, …)



Čárové spektrum

tvořené úzkými, navzájem oddělenými spektrálními čárami o různé intenzitě, zdroj: výboj v plynu za sníženého tlaku jiskrový výboj.

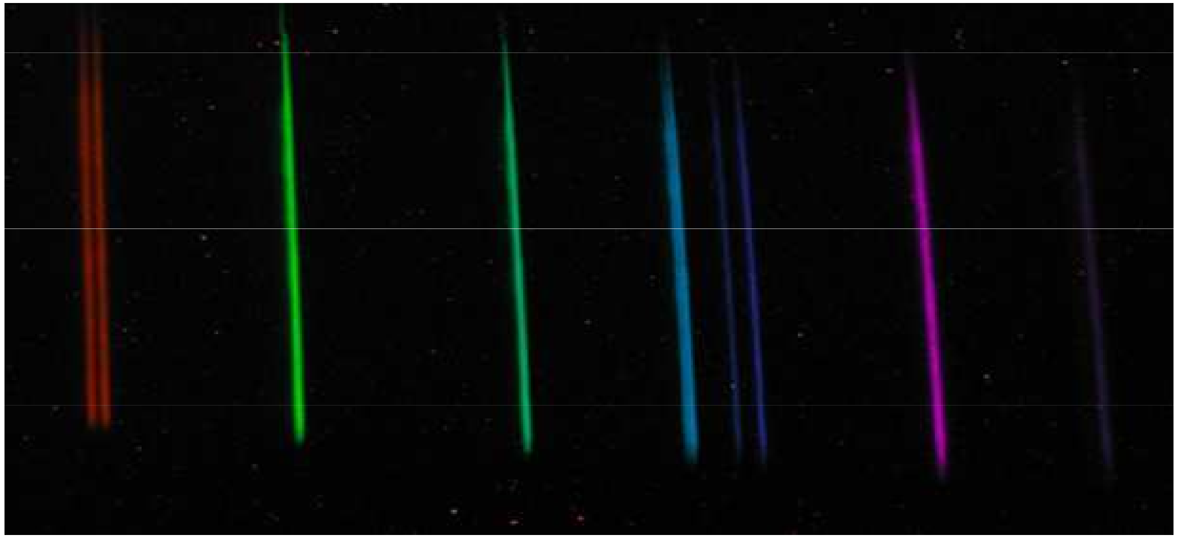
emisní spektrum sodíku



Pásové spektrum

tvořené pásy s množstvím spektrálních čar těsné blízkosti, mezi nimiž jsou temné úseky, zdroj: zářící molekuly látek.

emisní spektrum směsi par kadmia, rtuti a zinku



Lambert – Beerův zákon

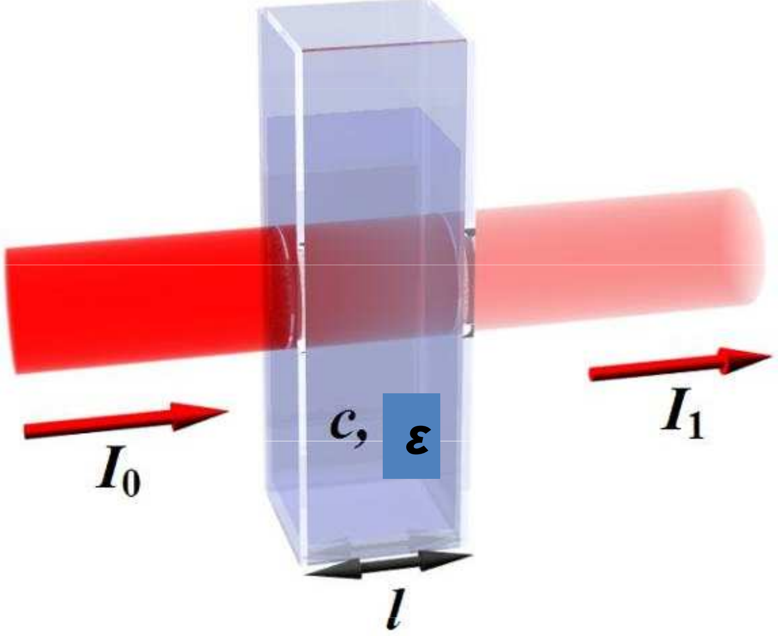
$$A = \log_{10}\frac{I_o}{I} = \varepsilon l c$$

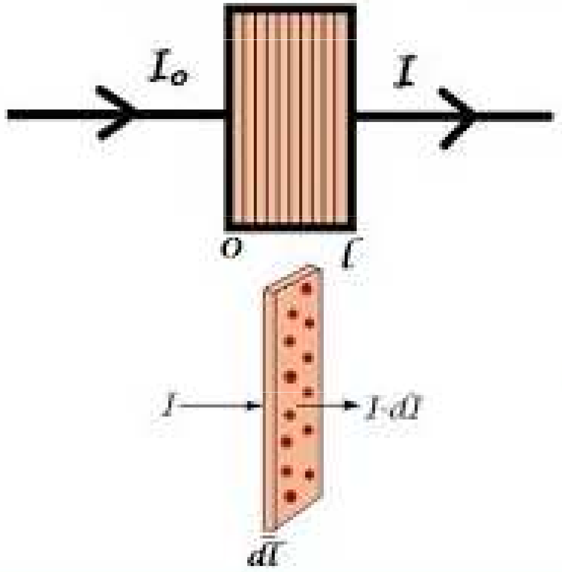
Zákon platí pro monochromatické světlo

l = tloušťka kyvety

c = koncentrace roztoku

ε = absorpční koeficient, ε = f(λ)





$$-\frac{dI}{dl} \propto cI$$

$$-\frac{dI}{I} = kc\,dl$$

$$\boxed{A = \epsilon c l}$$

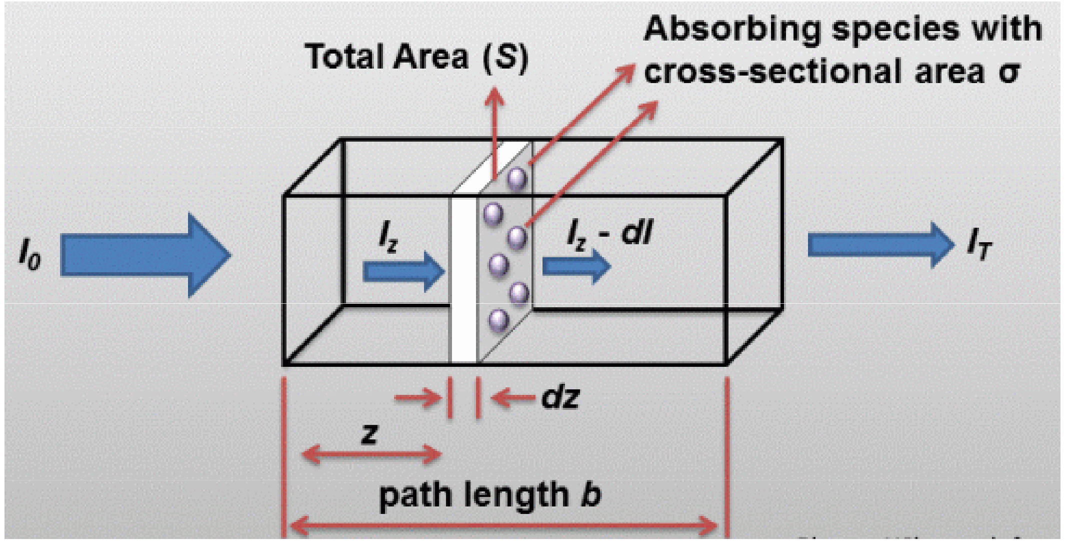
$$T(\%) = 100\frac{I}{I_0}$$

T = transmitance

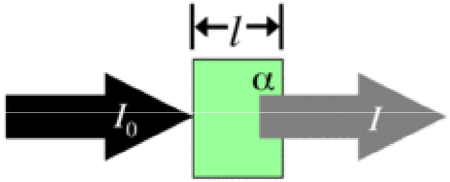
$$A = \log_{10}\frac{I_0}{I}$$

$$A = -\log_{10} T$$

A = absorbance

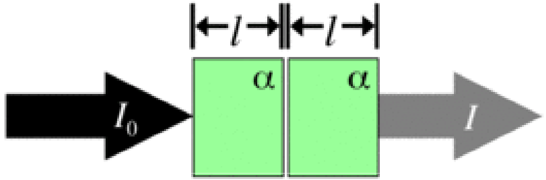


If *one slab* of absorbing material of thickness *l* reduces the intensity of a beam of light to *half*.



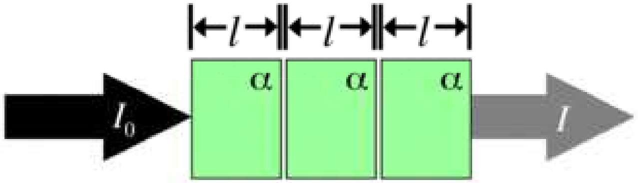
$$\frac{I}{I_0} = 10^{-kl} = \frac{1}{2}$$

Then *two slabs* of the same absorbing material will then reduce the intensity of a beam of light to *one quarter*.

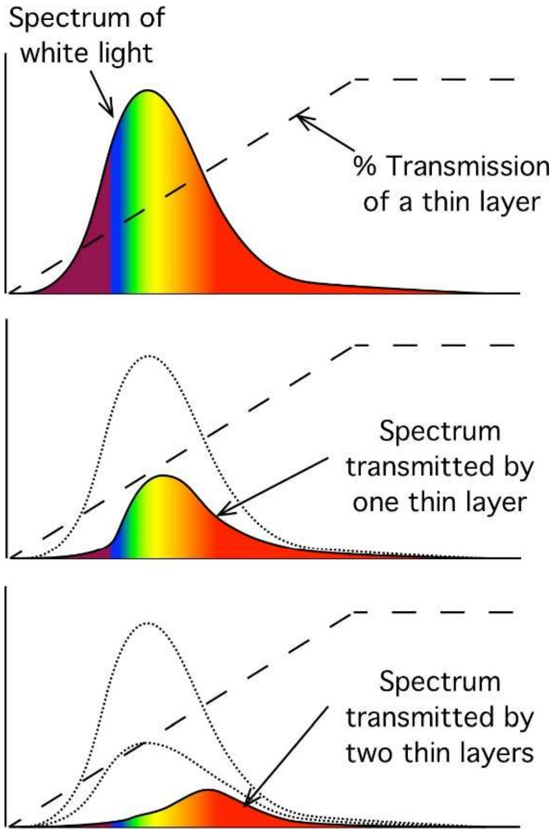


$$\frac{I}{I_0} = 10^{-k2l} = \left(\frac{1}{2}\right)^2 = \frac{1}{4}$$

And *three slabs* will reduce the intensity of a beam of light to *one eight*.

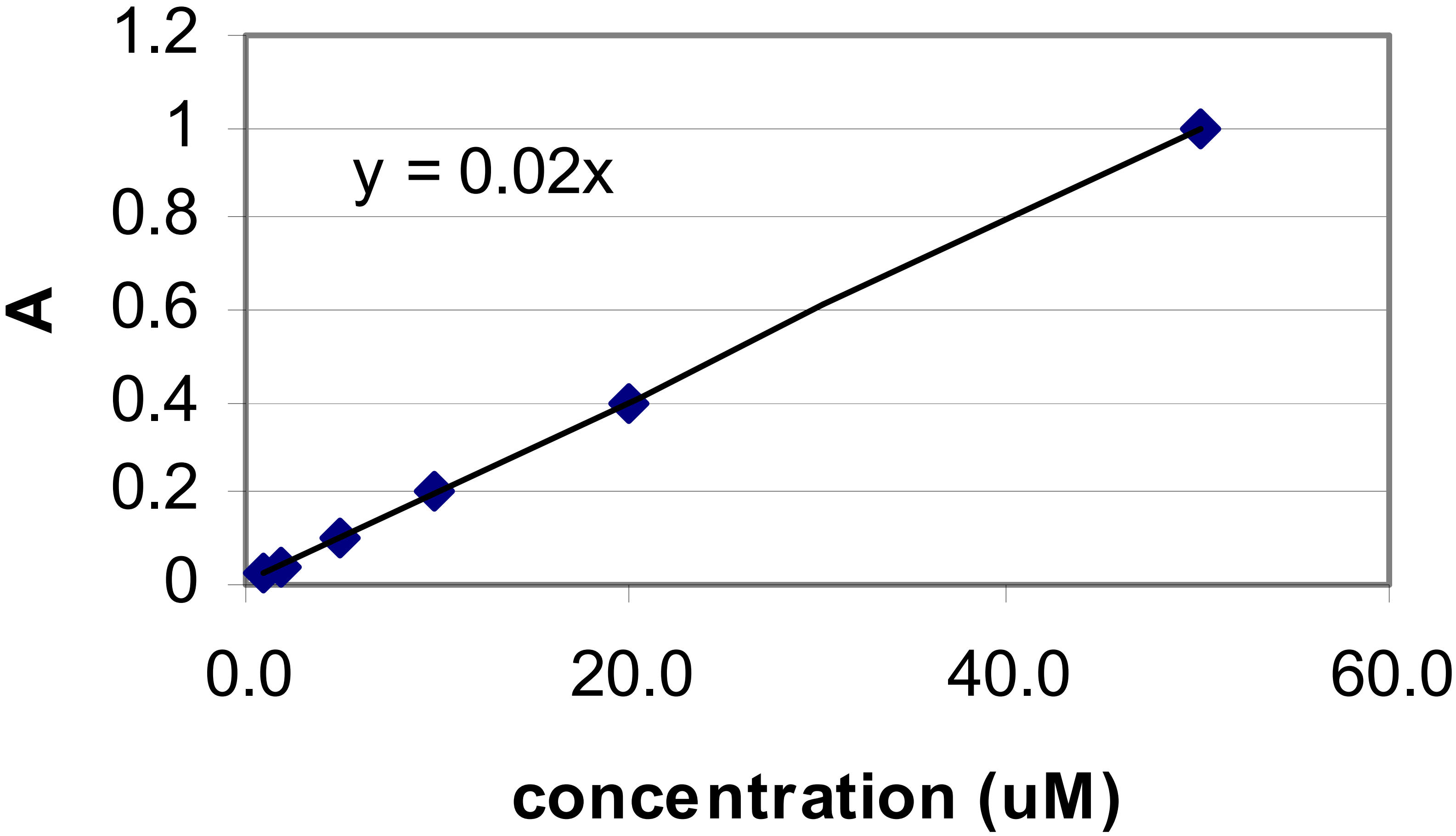


$$\frac{I}{I_0} = 10^{-k3l} = \left(\frac{1}{2}\right)^3 = \frac{1}{8}$$

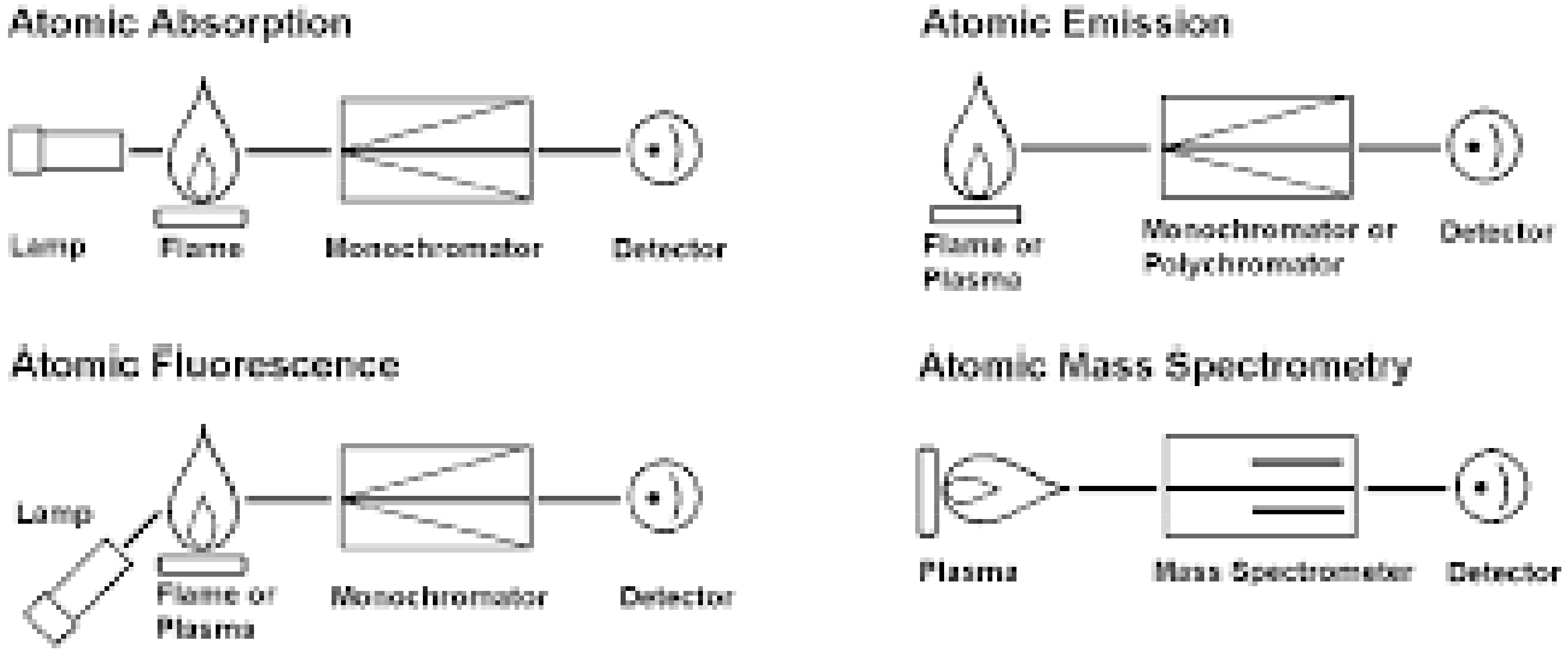


Grafické vyjádření Lambert-Beerova zákona

= lineární funkce



Spektrometrie



Spectroscopic Techniques and Chemistry they Probe

UV-Vis	UV-vis region	bonding electrons
Atomic Absorption	UV-vis region	atomic transitions (val. e-)
FT-IR	IR/Microwave	vibrations, rotations
Raman	IR/UV	vibrations
FT-NMR	Radio waves	nuclear spin states
X-Ray Spectroscopy	X-rays	inner electrons, elemental
X-ray Crystallography	X-rays	3-D structure

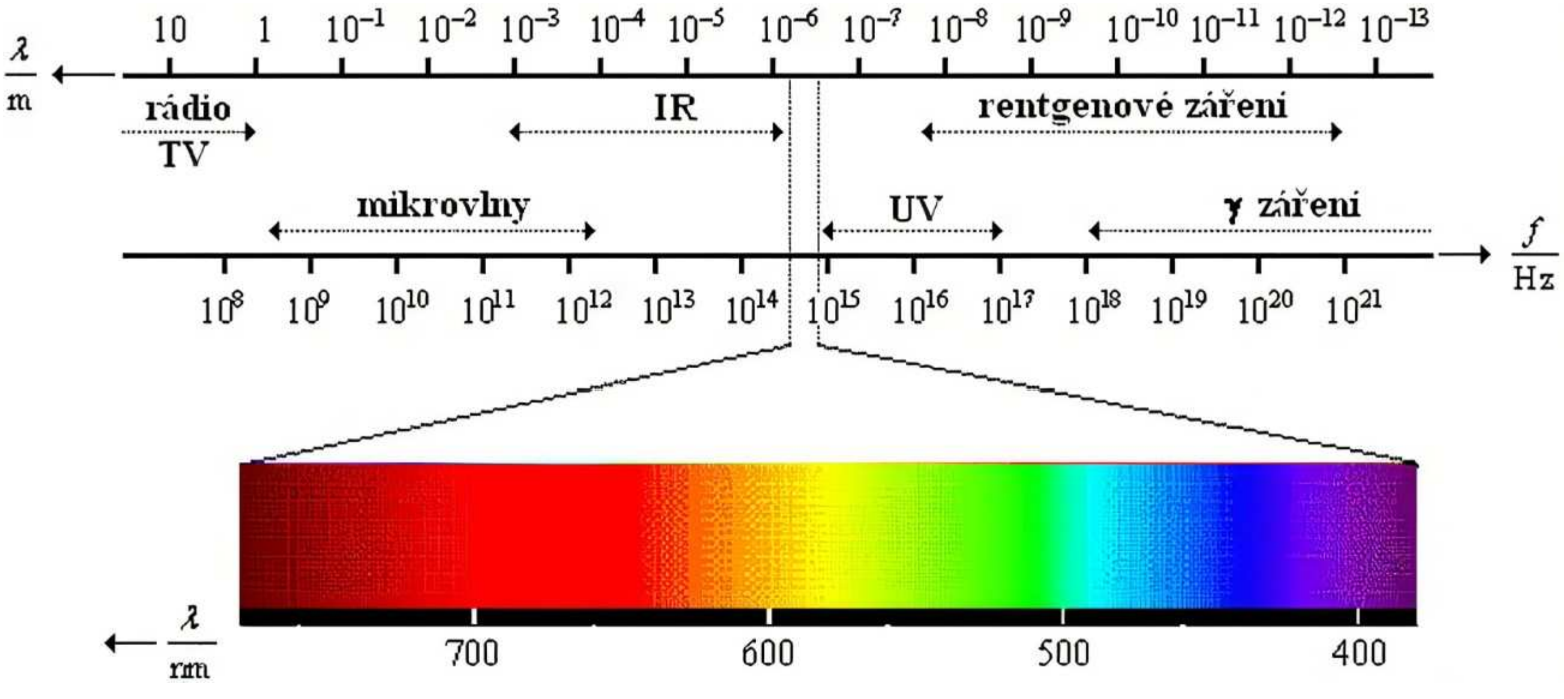
Spectroscopic Techniques and Common Uses

UV-vis	UV-vis region	Quantitative analysis/Beer’s Law
Atomic Absorption	UV-vis region	Quantitative analysis Beer’s Law
FT-IR	IR/Microwave	Functional Group Analysis
Raman	IR/UV	Functional Group Analysis/quant
FT-NMR	Radio waves	Structure determination
X-Ray Spectroscopy	X-rays	Elemental Analysis
X-ray Crystallography	X-rays	3-D structure Anaylysis

UV-VIS

Světlo (viditelné záření) je součástí spektra elektromagnetického záření, lidské oko vnímá elektromagnetické vlnění o frekvencích $7,6 \cdot 10^{14}$ Hz – $3,9 \cdot 10^{14}$ Hz.

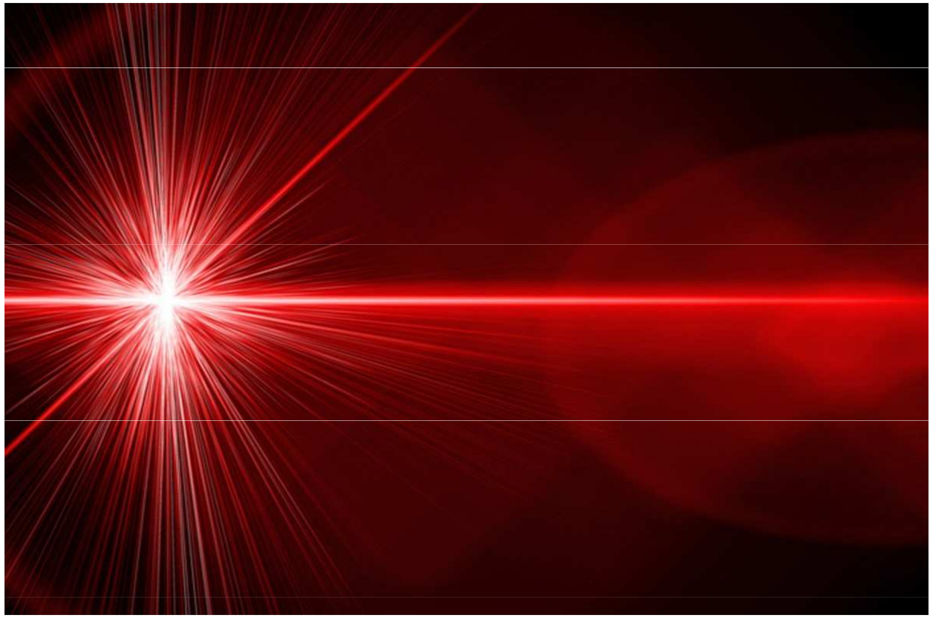
Bílé světlo



Světelné spektrum je část elektromagnetického spektra, ve kterém je zobrazena závislost barev světla na vlnových délkách.

Chromatické světlo: složené ze světla více vlnových délek, např. bílé světlo (složené ze sedmi barev)

Monochromatické světlo: pouze jedna vlnová délka, např. laser



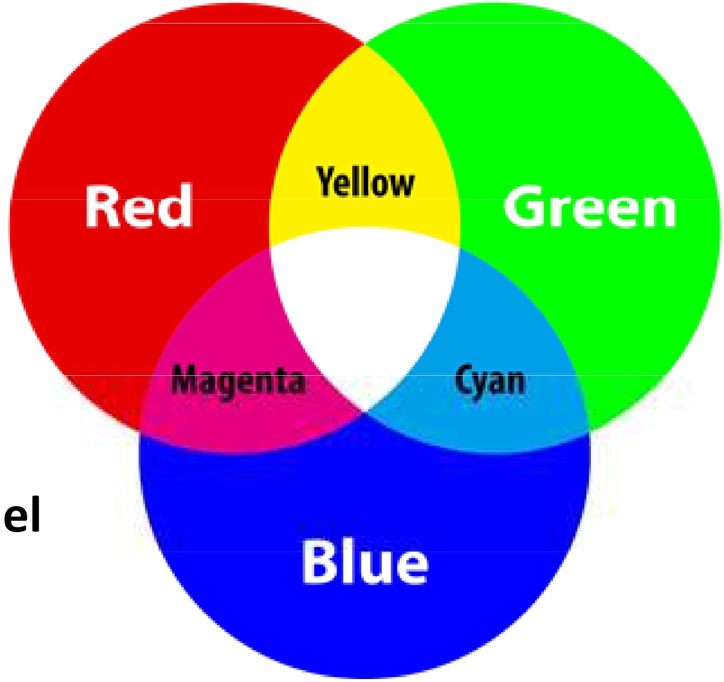
Míchání barev

Aditivní (součtové) - RGB

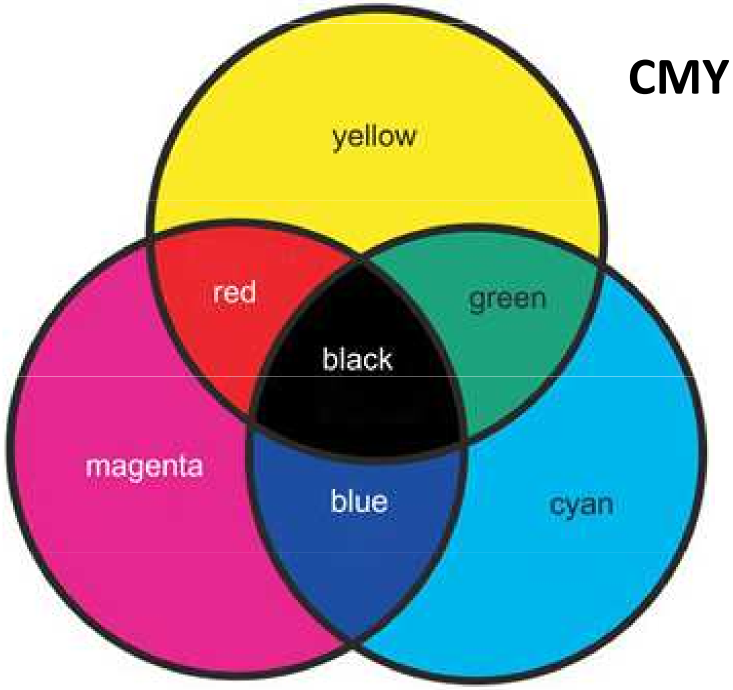
Jednotlivé složky barev se sčítají a vytváří světlo větší intenzity. Smícháním dvou základních barev vznikne **komplementární** (**doplňková**) barva k třetí základní barvě. Princip aditivního míchání barev se uplatňuje na počítačových monitorech a TV obrazovkách.

Subtraktivní (odčítací) CMYK

S každou další přidanou barvou se ubírá část původního světla. Využití v barevných tiskárnách.



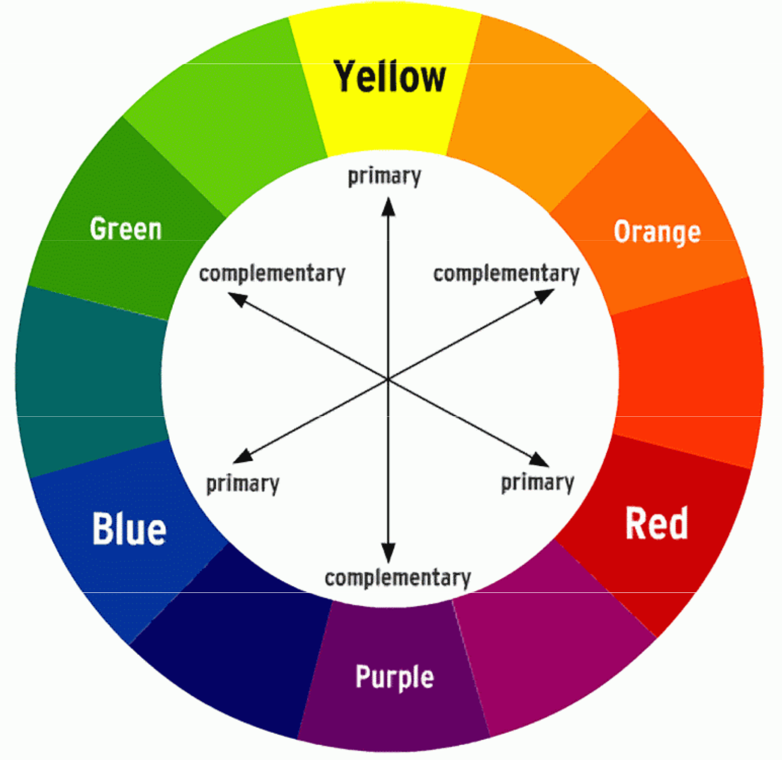
RGB model

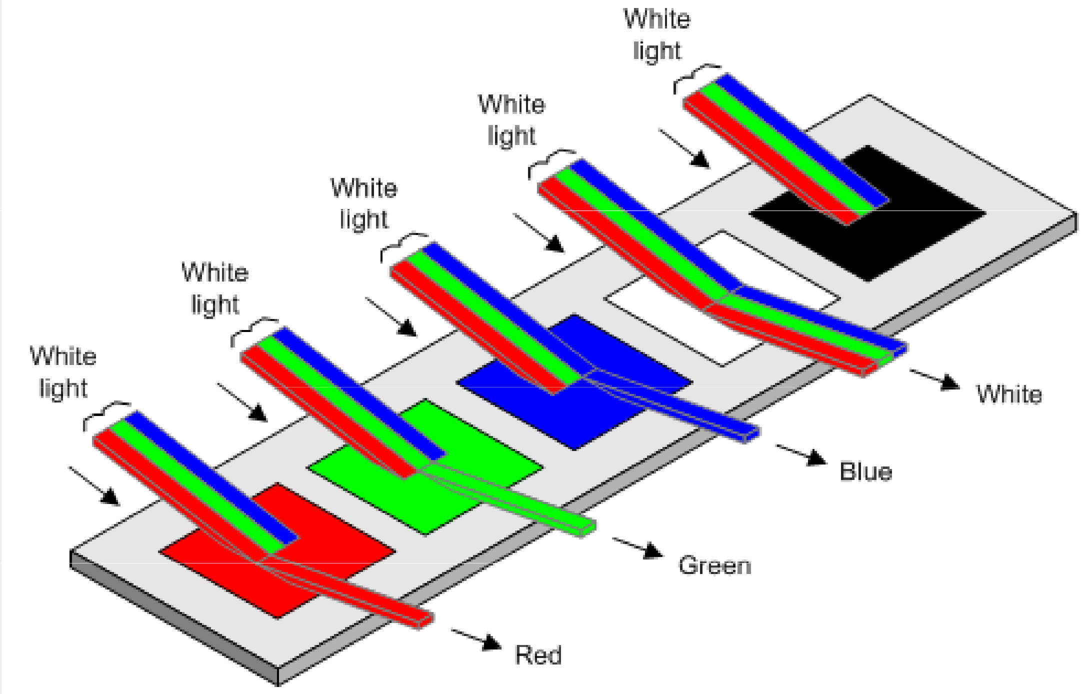


CMYK model

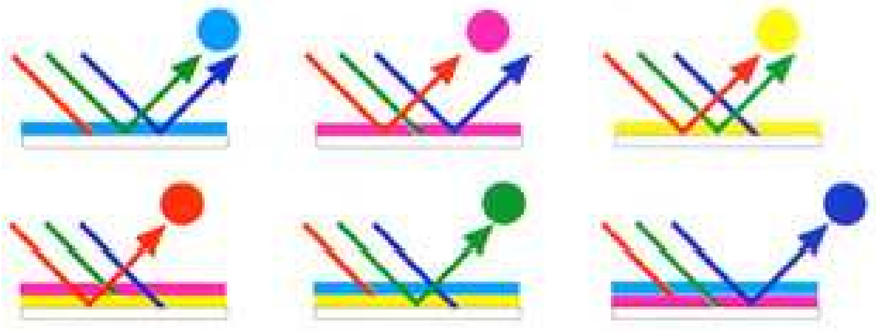
Absorpce záření

- odražené světlo = barva předmětu
- černé těleso vše pohlcuje, bílé vše odráží

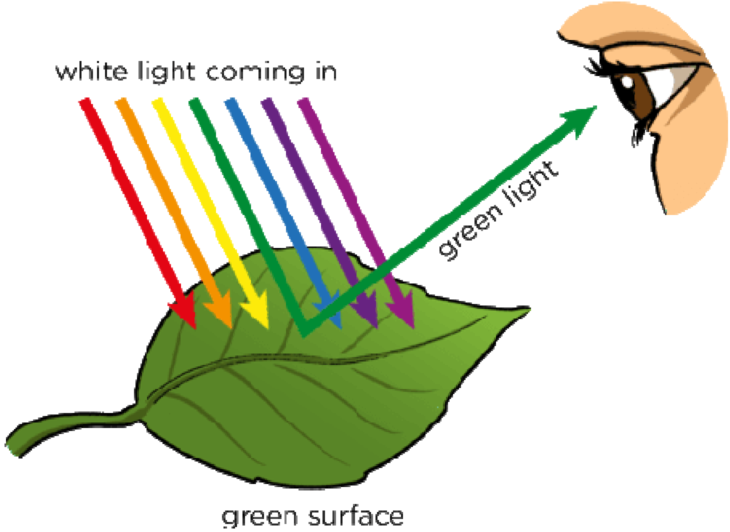


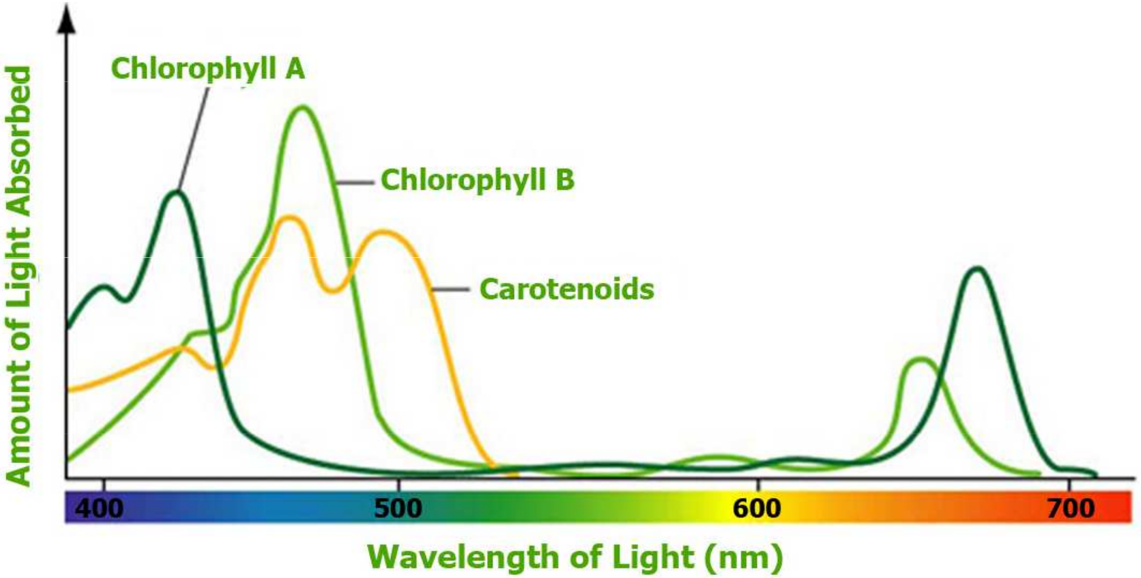


Shining white light on different colored paints

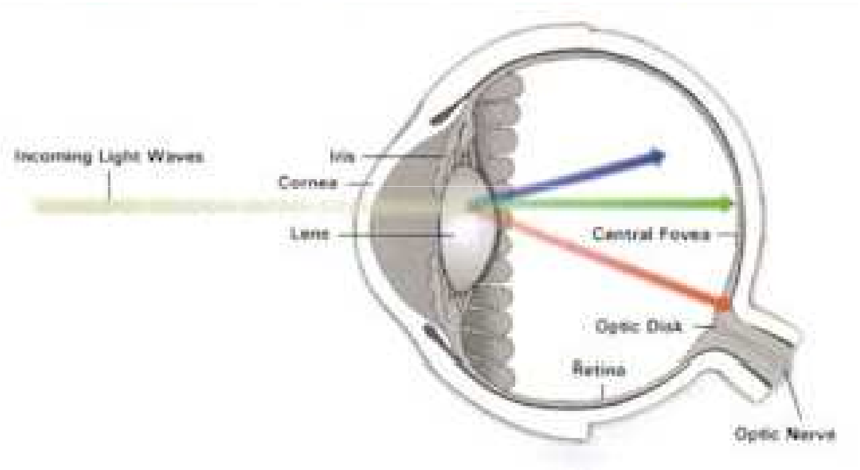


Příklad


white light coming in
green light
green surface

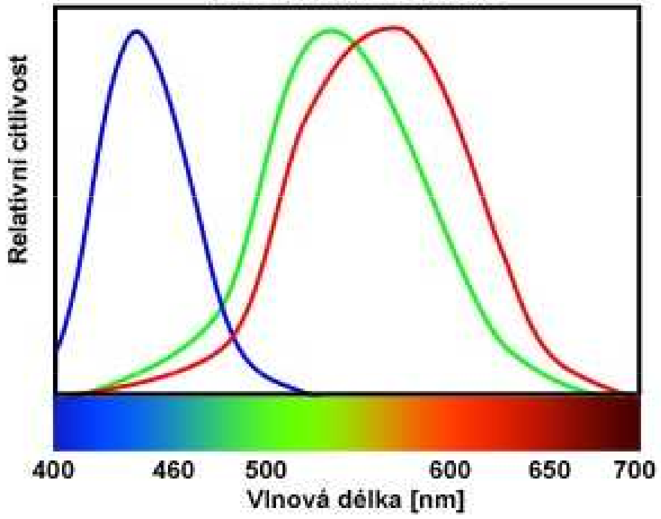

Chlorophyll A
Chlorophyll B
Carotenoids
Amount of Light Absorbed
400
500
600
700
Wavelength of Light (nm)

Lidské oko

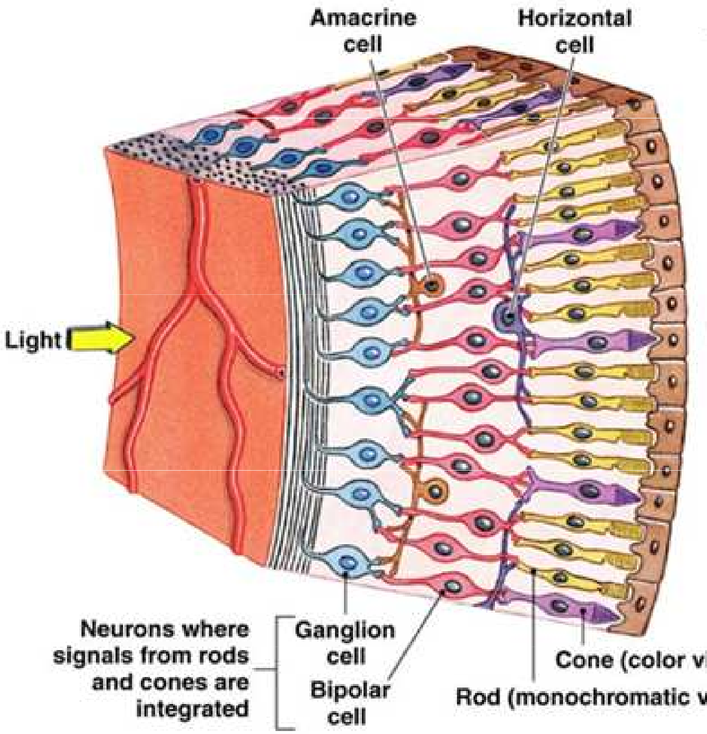


Citlivost lidského oka na barvy

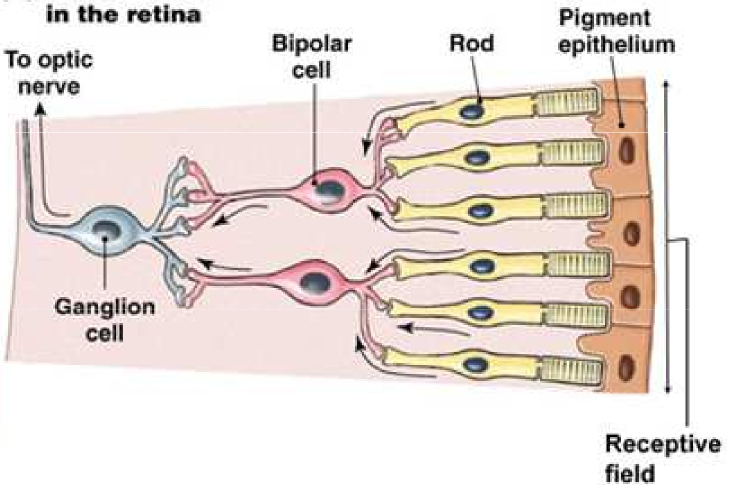
3 druhy buněk oka hrubě odpovídají modré, zelené a červené

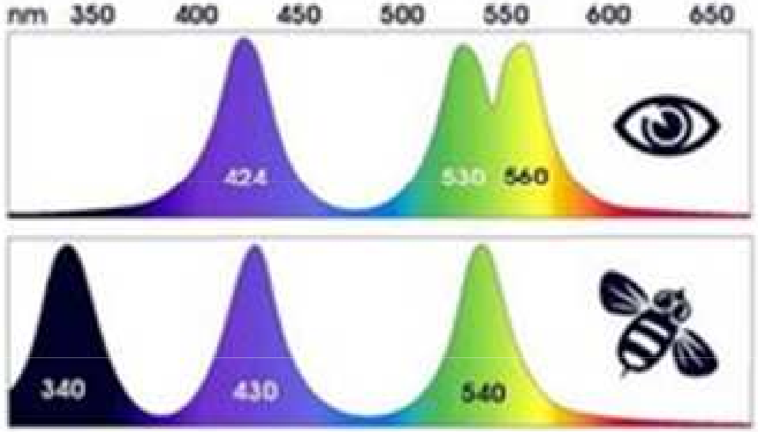


(d) Organization of the retina

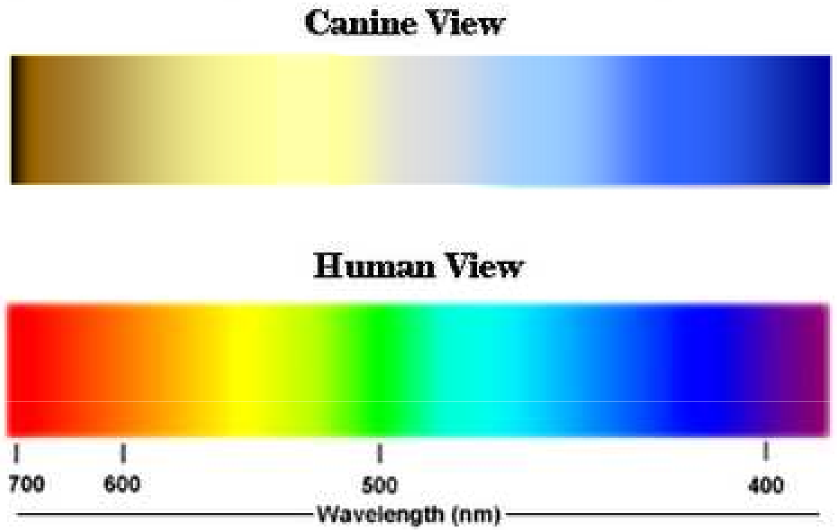


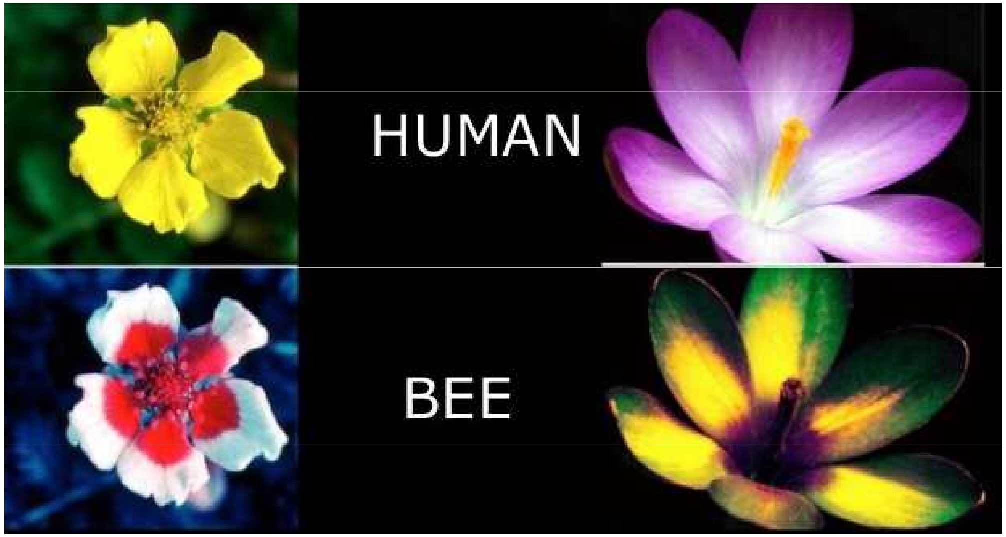
(e) Convergence in the retina





How do dogs and human see spectrum





How we see

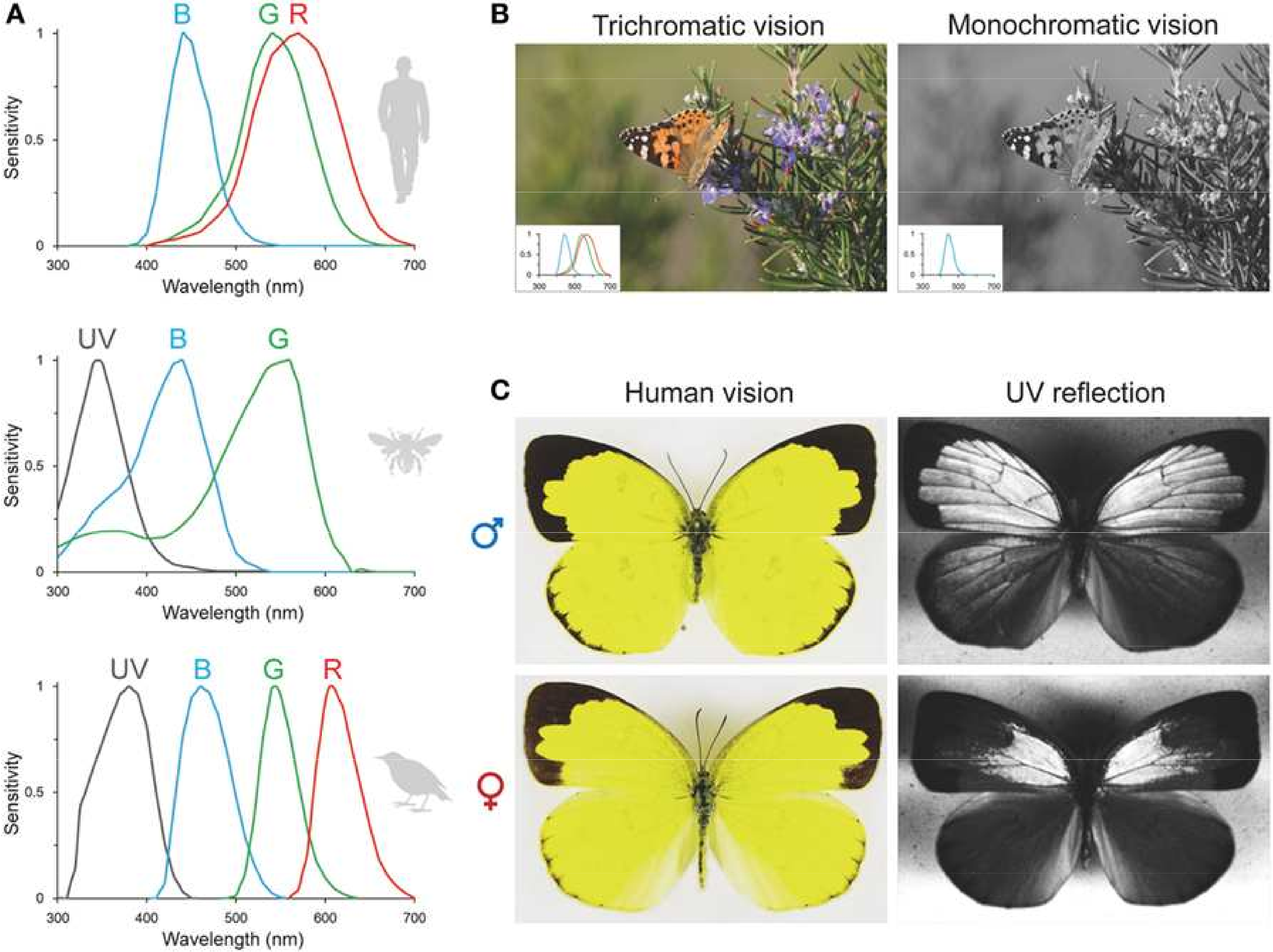
Colours: Red, Green & Blue

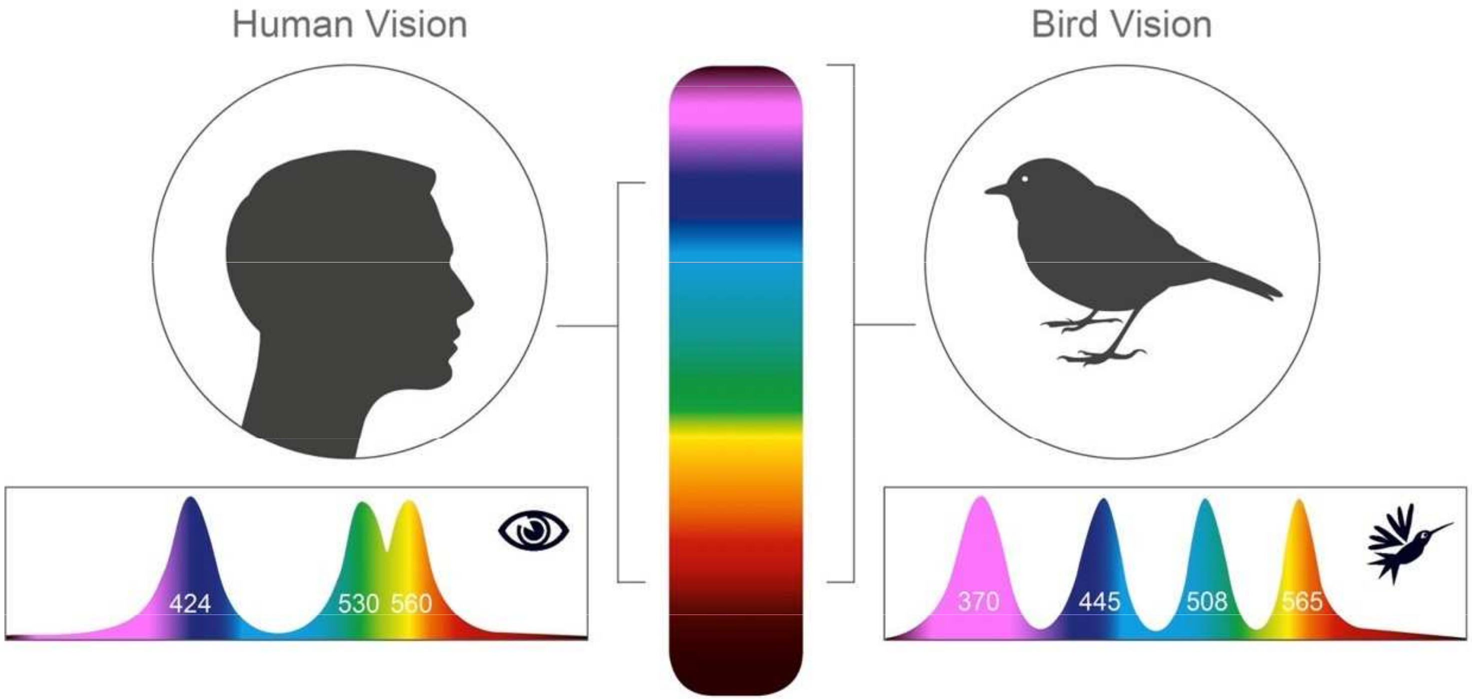
We see all other colours in combination of these 3 colours

How dogs see

Colours: Blue & Yellow (can be compared to the vision of a colour-blind human)

Have a wider peripheral vision

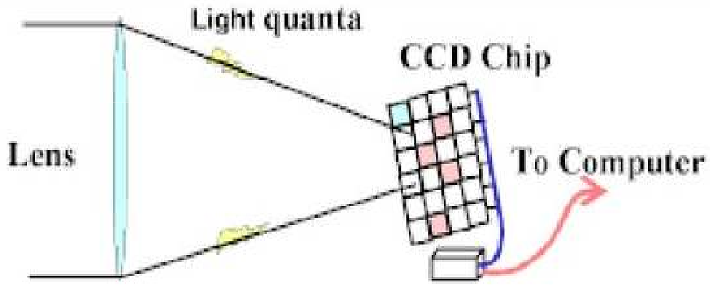

A
B
G
R
Sensitivity
1
0.5
0
300
400
500
600
700
Wavelength (nm)
UV
B
G
Sensitivity
1
0.5
0
300
400
500
600
700
Wavelength (nm)
UV
B
G
R
Sensitivity
1
0.5
0
300
400
500
600
700
Wavelength (nm)
B
Trichromatic vision
Monochromatic vision
1
0.5
0
300
500
700
C
Human vision
UV reflection
♂
♀

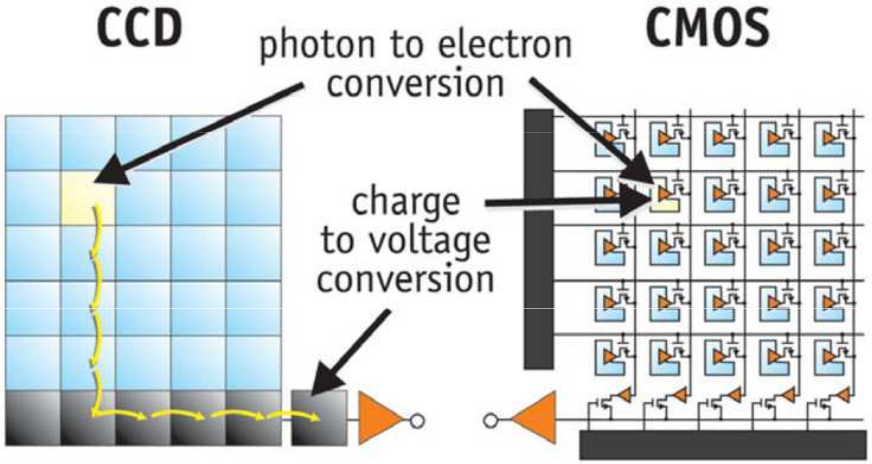

Human Vision
Bird Vision
424
530
560
370
445
508
565


Human vision
Bird vision

CCD a CMOS detektory

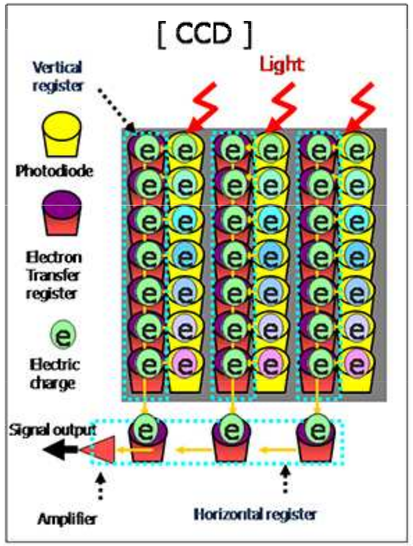
U **CCD** je nakumulovaný náboj přesouván přes matici Schottkyho fotodiod. Elektrony reprezentující jednotlivé pixely jsou posouvány do výstupního zesilovače, kde je elektrický náboj převeden na napětí. Výhodami tohoto detektoru oproti typu CMOS je lepší světelná citlivost, což se projeví v lepší kvalitě obrazu při špatném osvětlení. CCD senzory také dosahují vyšší rychlosti převodu signálu a výsledný obraz vykazuje relativně nízký šum. Nevýhodou těchto detektorů je vyšší cena a je složitější instalace do kamery.

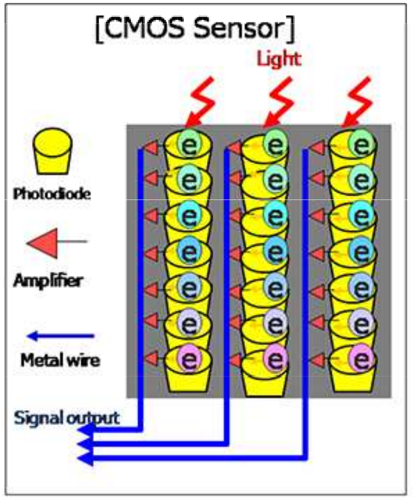




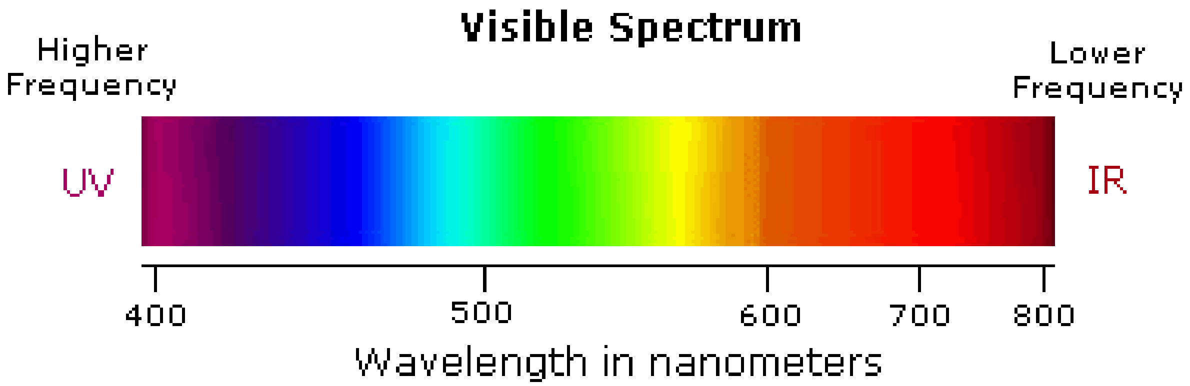
CCDs move photogenerated charge from pixel to pixel and convert it to voltage at an output node. CMOS imagers convert charge to voltage inside each pixel.

CMOS využívá integrovaných obvodů vysoké hustoty, umožňující umístit na čip velké množství MOS tranzistorů. Produkce těchto detektorů je sériová a levnější než u CCD prvků. Výhodou CMOS senzorů je také nízká spotřeba energie, nižší napájecí napětí a obecně jednodušší elektronika, umožňující návrh kamer s menšími rozměry.

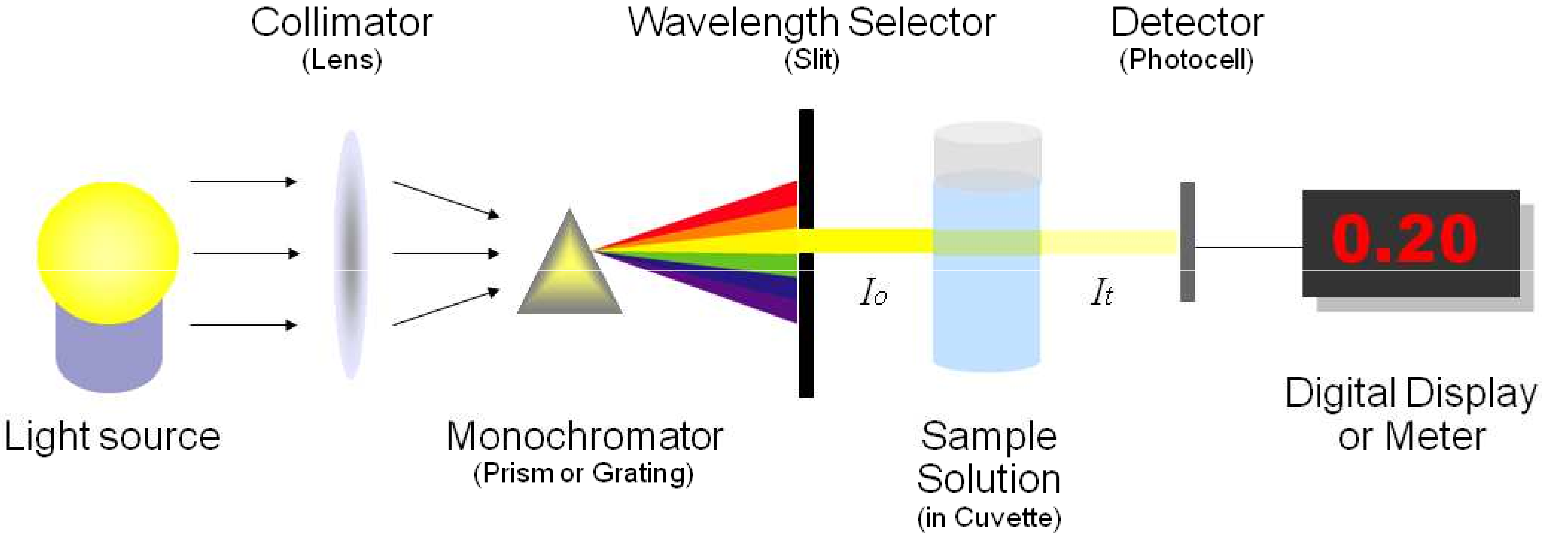




UV-VIS spektrometrie

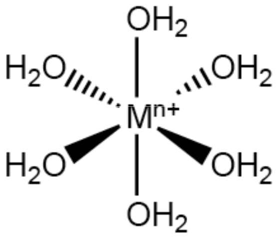


Violet: 400 - 420 nm
Indigo: 420 - 440 nm
Blue: 440 - 490 nm
Green: 490 - 570 nm
Yellow: 570 - 585 nm
Orange: 585 - 620 nm
Red: 620 - 780 nm



TRANSITION METAL ION COLOURS

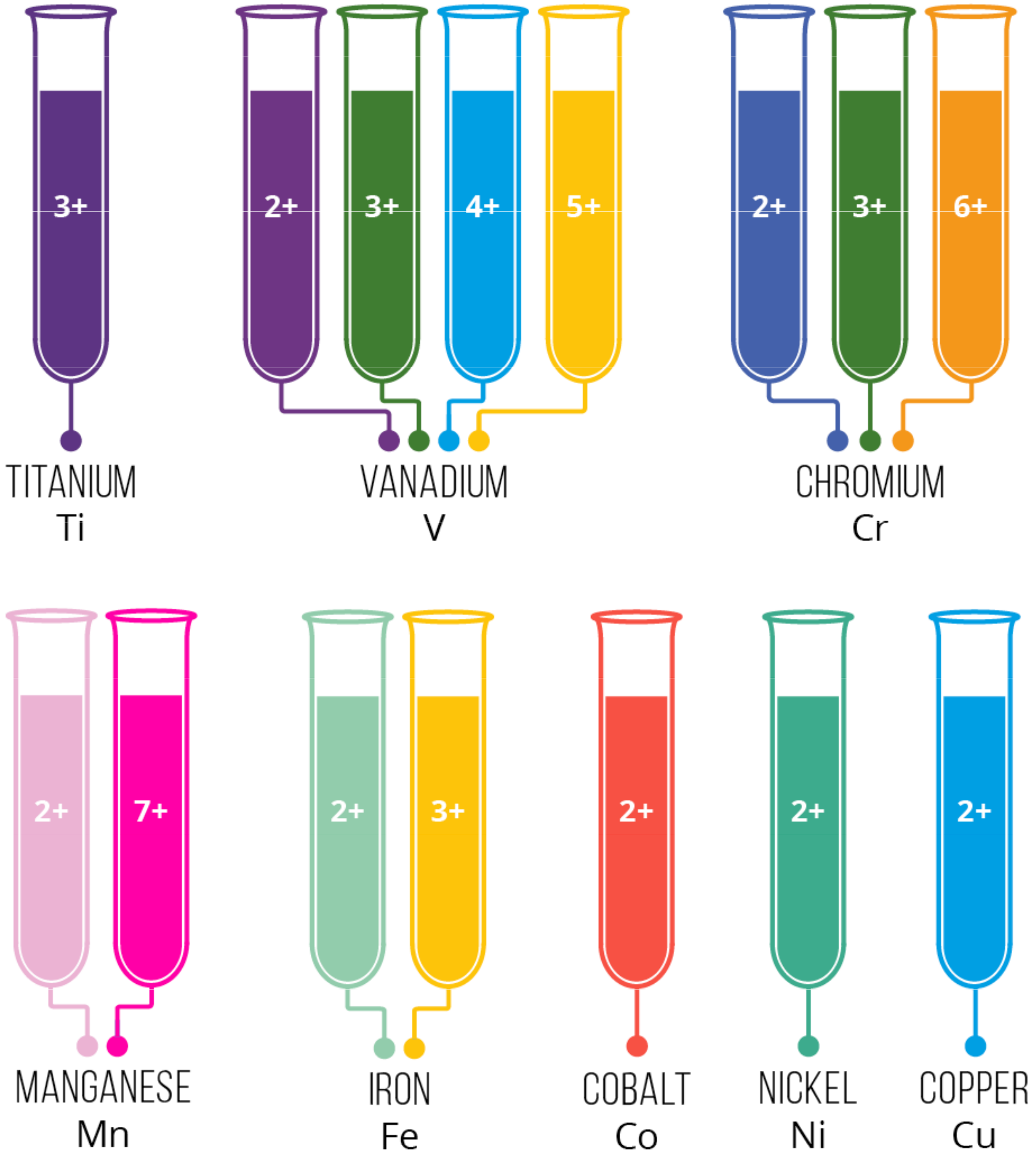
Transition metals form coloured compounds and complexes. These colours can vary depending on the charge on the metal ion, and the number and type of groups of atoms (called ligands) attached to the metal ion. In aqueous solutions, the ions form complexes with the colours shown to the right.

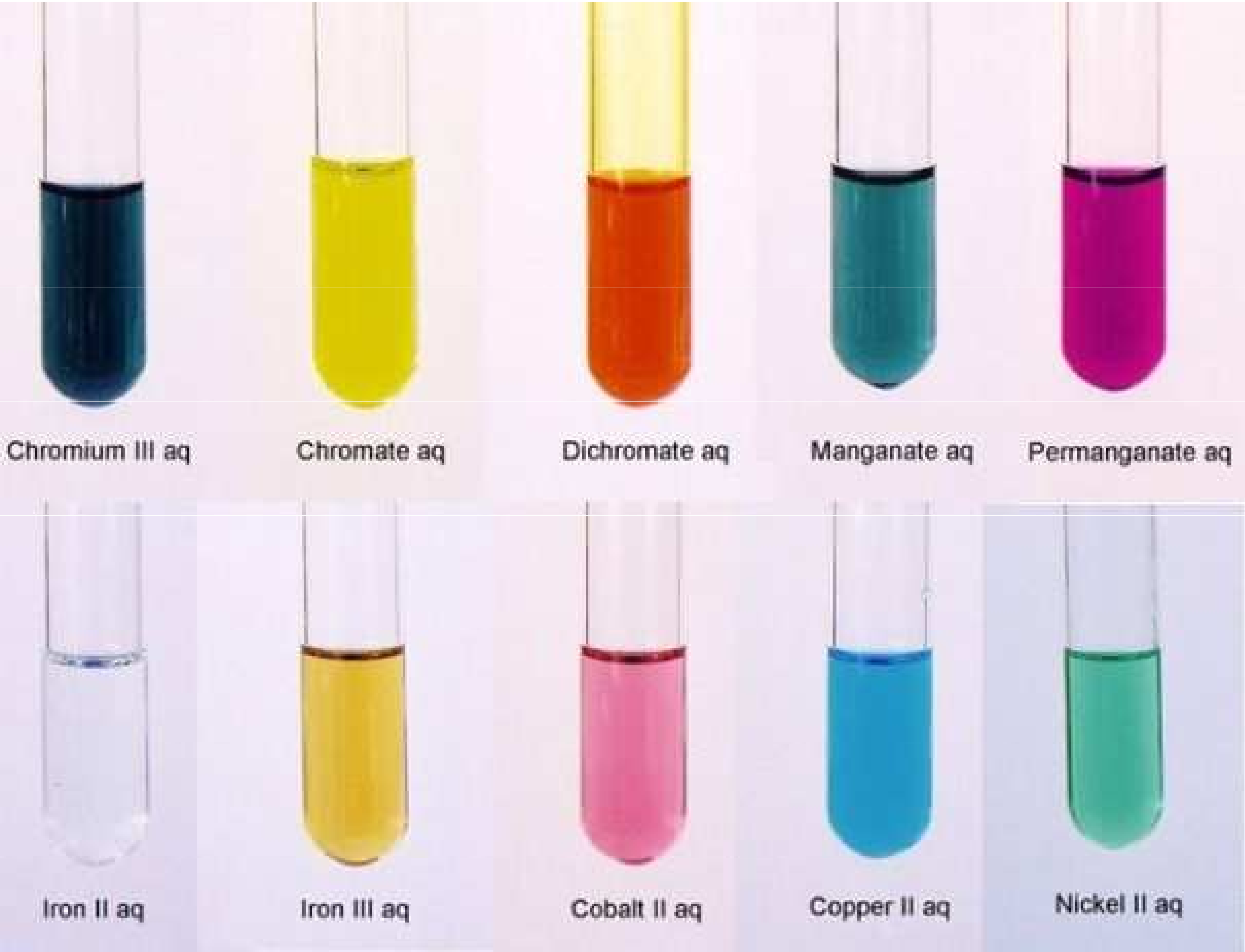


HYDRATED TRANSITION METAL ION

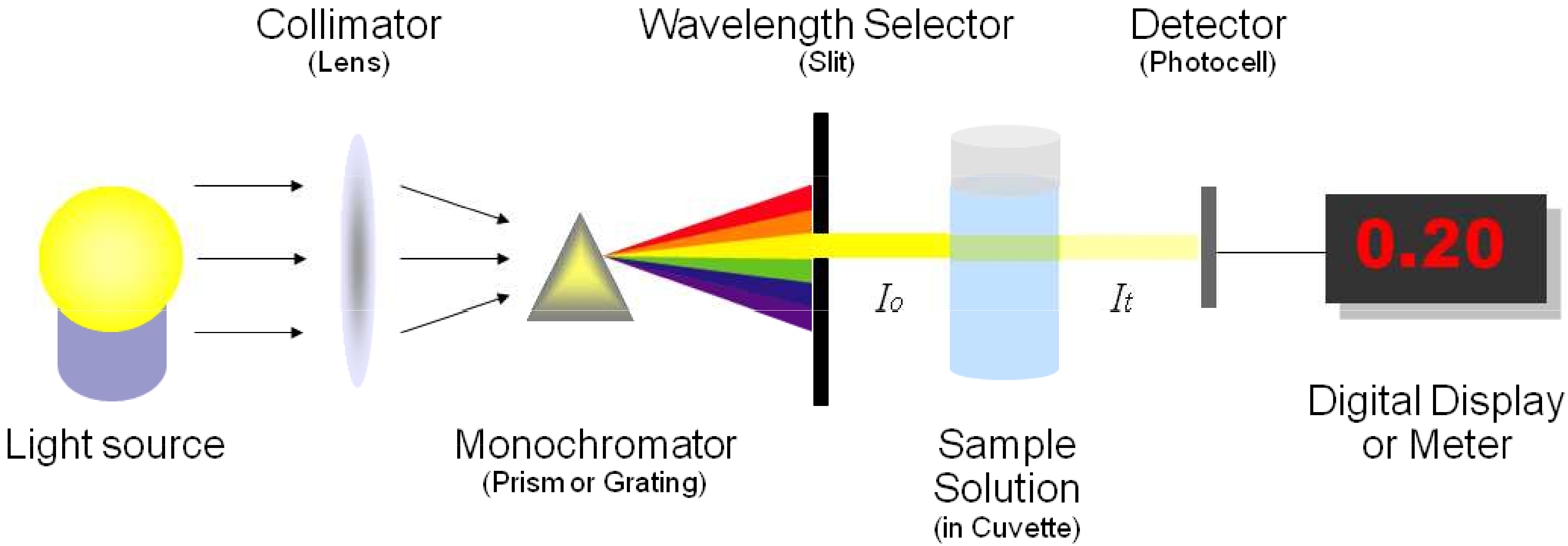
Electrons are arranged around the nucleus of the metal atom in orbitals. Transition metals, unlike other metals, have partially filled d orbitals, which can hold up to 10 electrons. When ligands are present, some d orbitals become higher in energy than before, and some become lower. Electrons can then move between these higher and lower d orbitals by absorbing a photon of light. This absorption of light affects the percieved colour of the compound or complex. The wavelength of the light absorbed is affected by the size of the energy gap between the d orbitals, which is in turn affected by the type of ligand and the charge on the metal ion.

2014 COMPOUND INTEREST WWW.COMPOUNDCHEM.COM

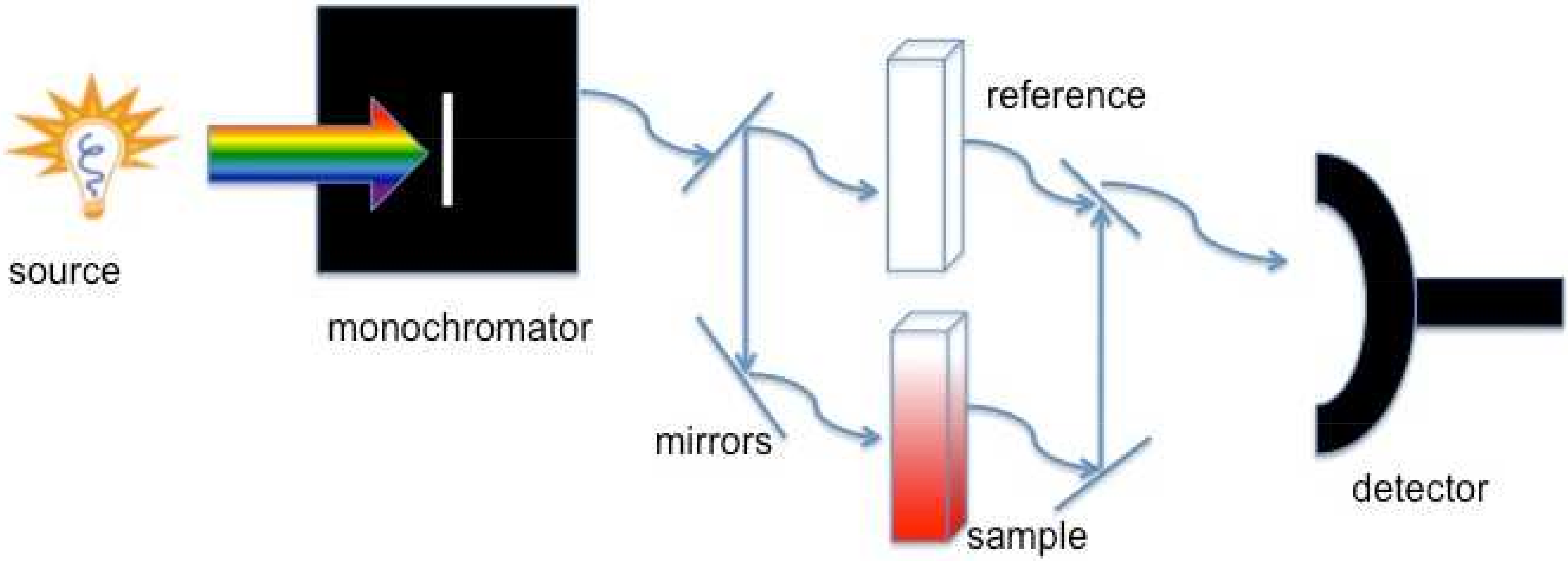



Chromium III aq
Chromate aq
Dichromate aq
Manganate aq
Permanganate aq
Iron II aq
Iron III aq
Cobalt II aq
Copper II aq
Nickel II aq

UV-Vis Spectrophotometer

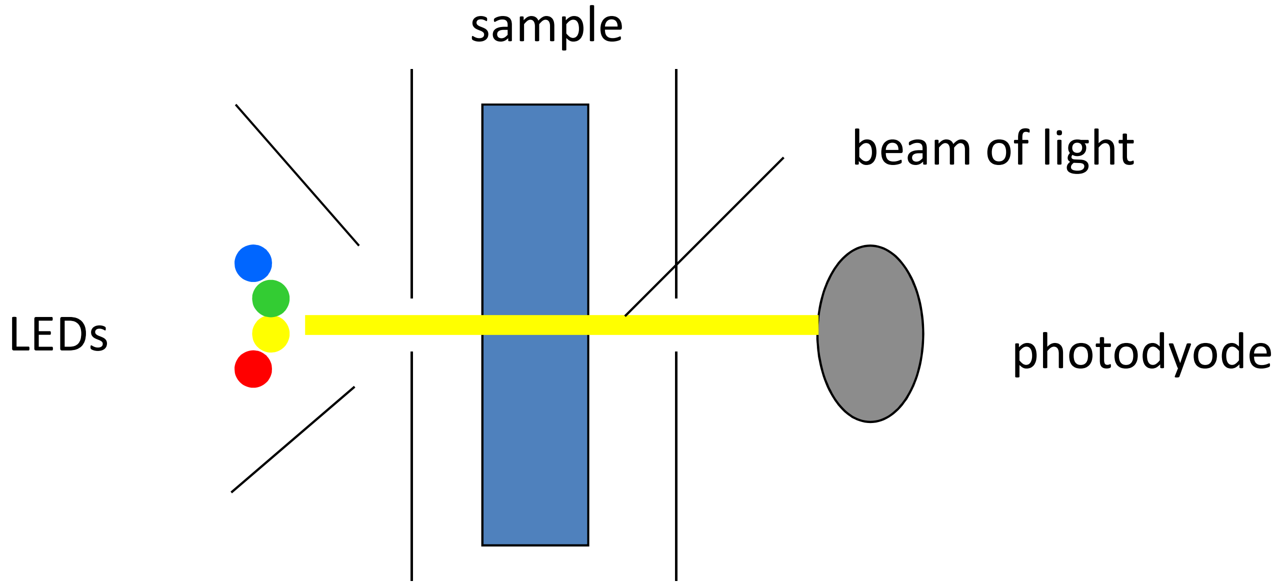


Double Beam UV-Vis Spectrophotometer

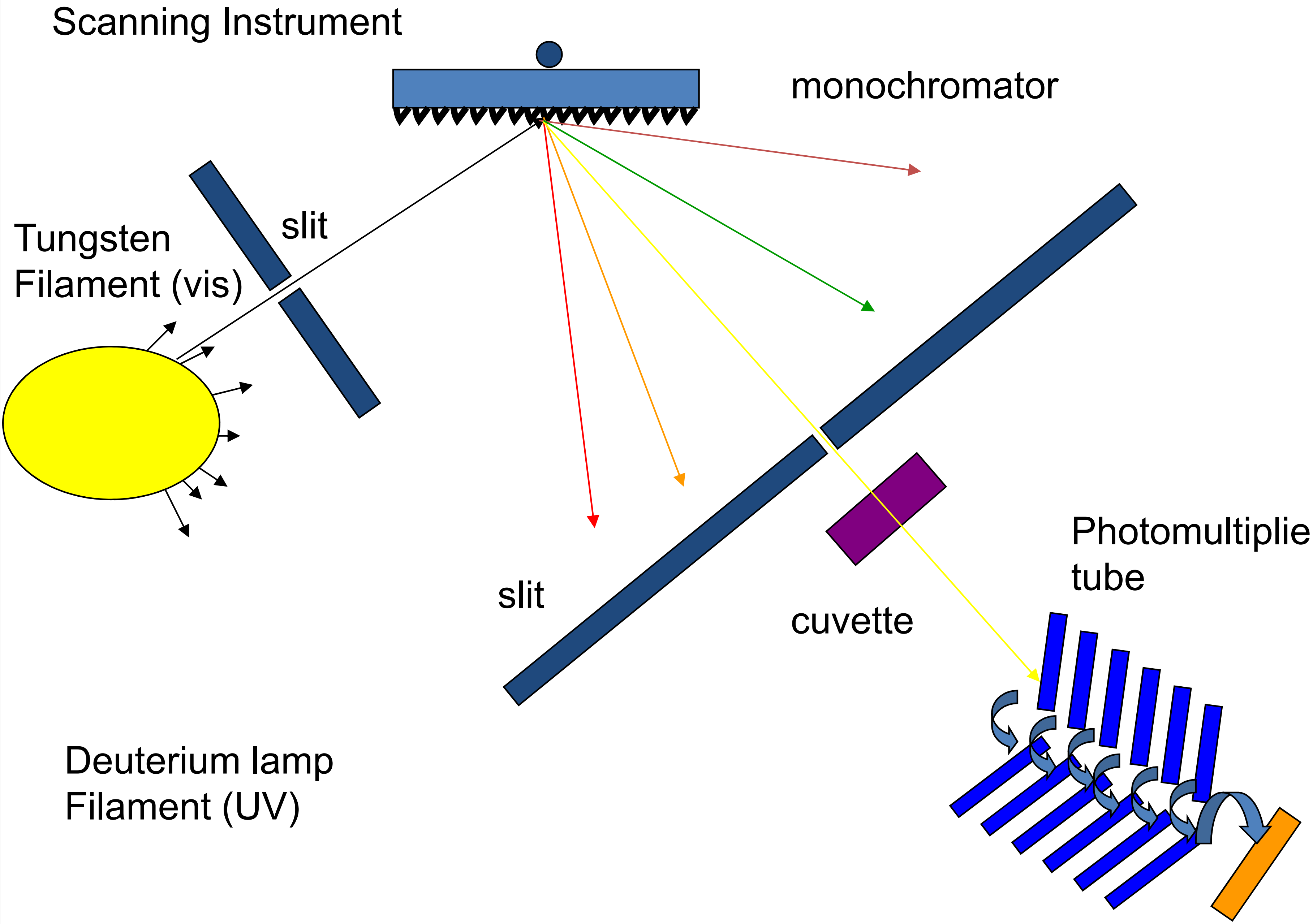


Fixed Wavelength Instrument

- LED serve as source
- Pseudo-monochromatic light source
- No monochrometer necessary/ wavelength selection occurs by turning on the appropriate LED
- 4 LEDs to choose from



Skenovací UV-VIS spektrometr



Scanning Instrument

monochromator

slit

Tungsten Filament (vis)

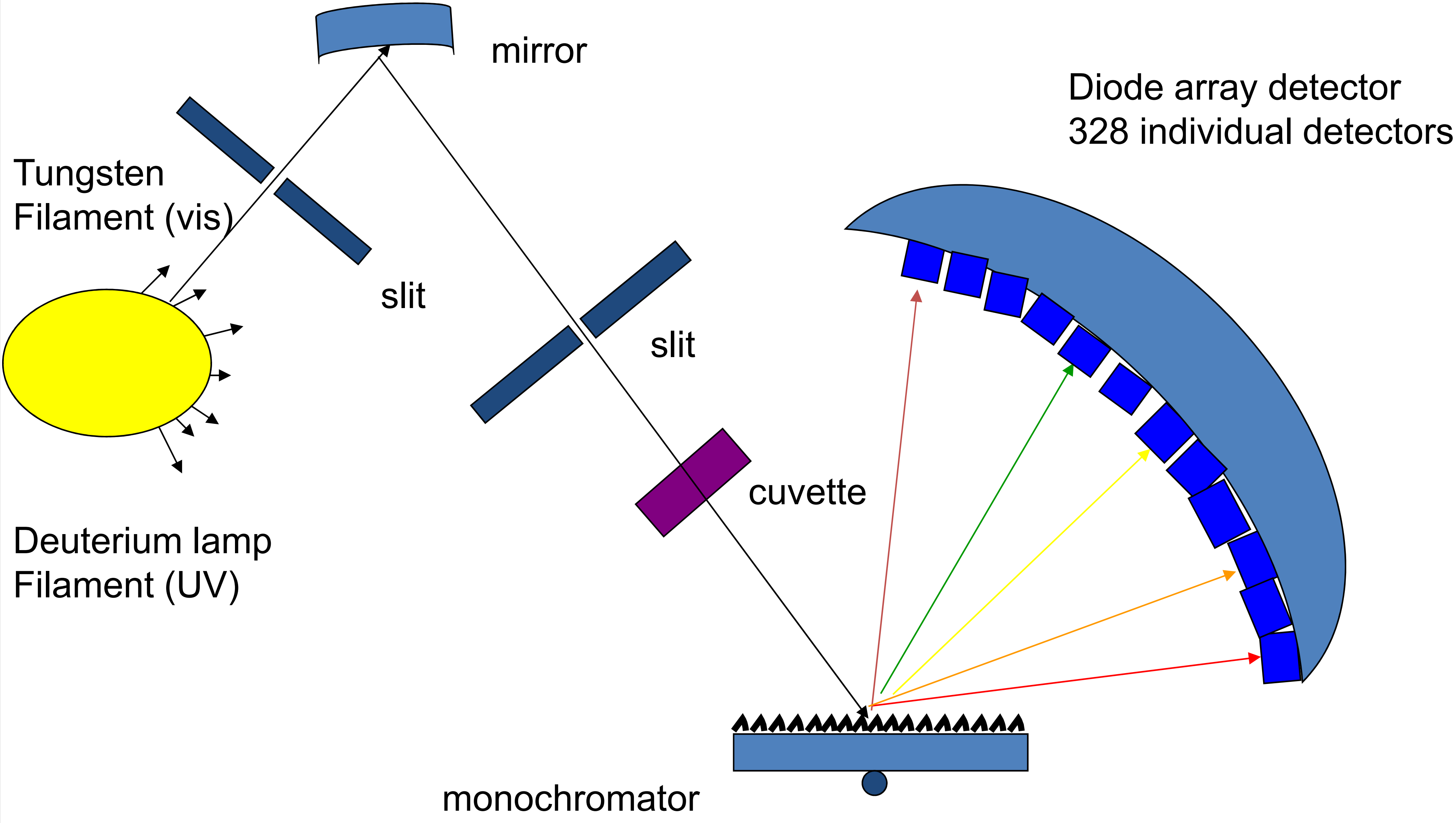
slit

cuvette

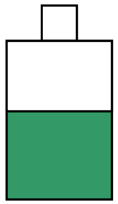
Photomultiplier tube

Deuterium lamp Filament (UV)

Diode array UV-VIS spektrometr

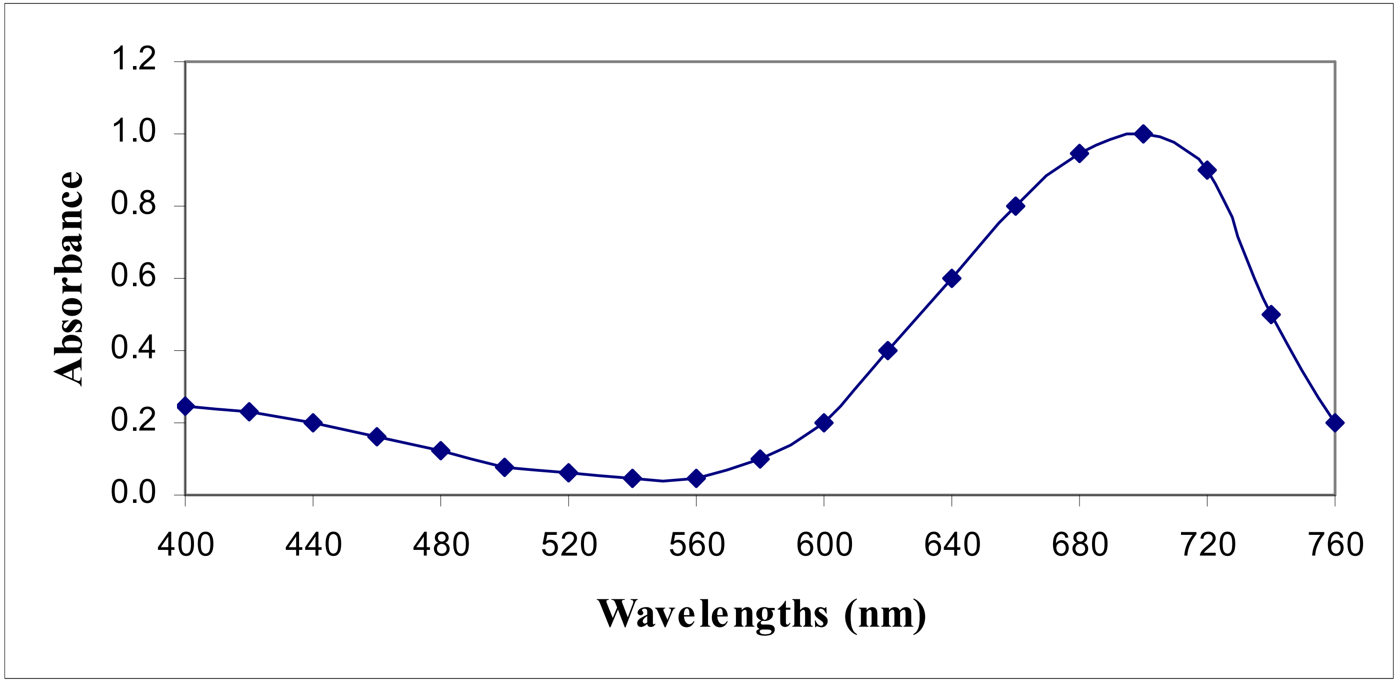


UV-Vis spectrum

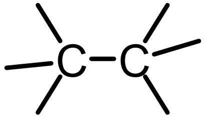


Sample Solution

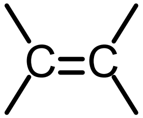
UV-Vis spectrum shows the absorbance of one or more sample components in the cuvette when scanned through various wavelengths



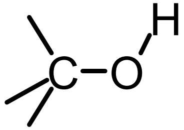
For the Solution first we'll determine where the maximum absorption occurs.



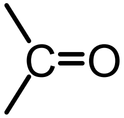
$\sigma \rightarrow \sigma^*$ 135 nm



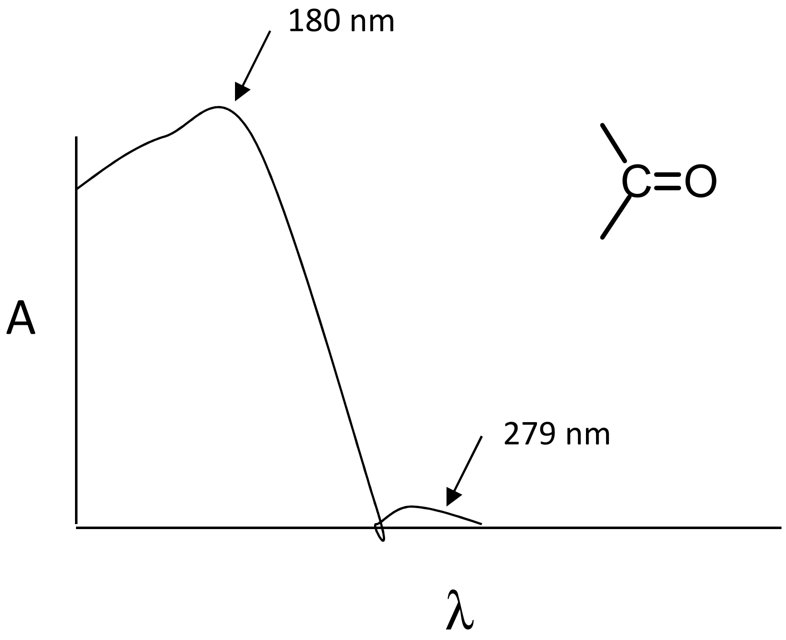
$\pi \rightarrow \pi^*$ 165 nm



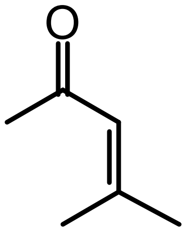
$n \rightarrow \sigma^*$ 183 nm weak



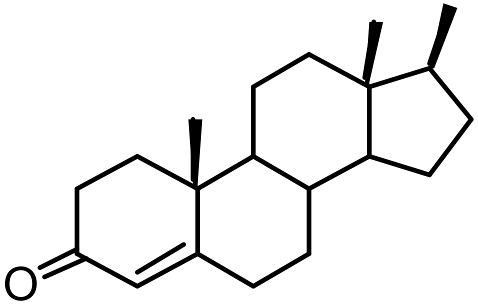
$\pi \rightarrow \pi^*$ 150 nm
$n \rightarrow \sigma^*$ 188 nm
$n \rightarrow \pi^*$ 279 nm weak



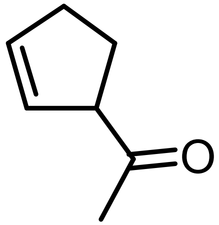
Similar structures have similar UV spectra:



λ_{max} = 238, 305 nm



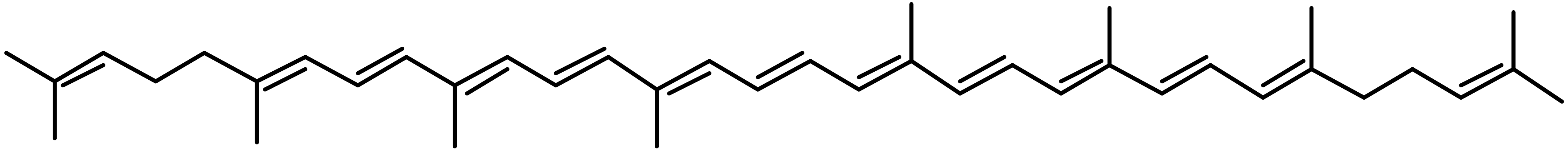
λ_{max} = 240, 311 nm



λ_{max} = 173, 192 nm

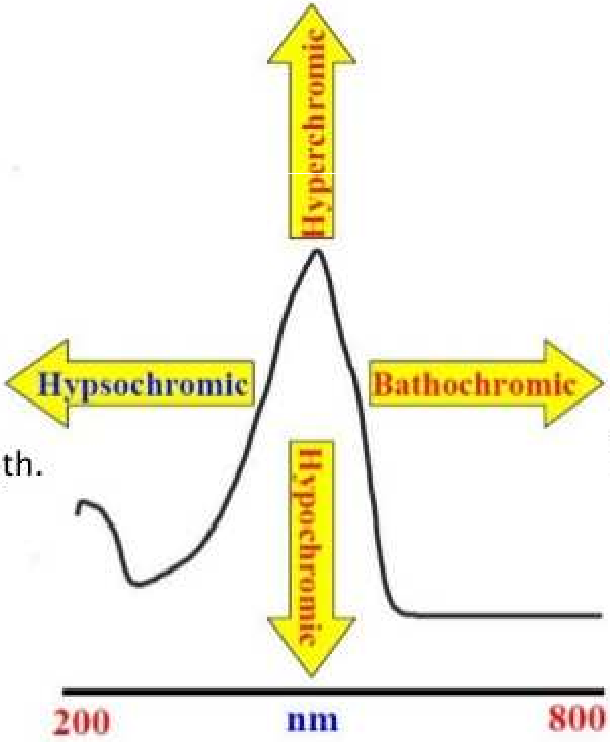
Barva souvisí s rozsahem konjugovaného systému

Lycopene:



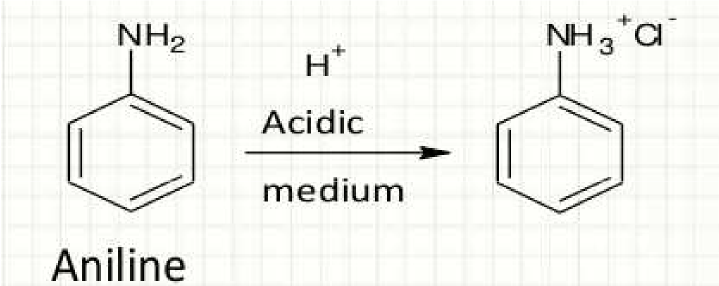
λ_{max} = 114 + 5(8) + 11*(48.0-1.7*11) = 476 nm

λ_{max}(Actual) = 474.



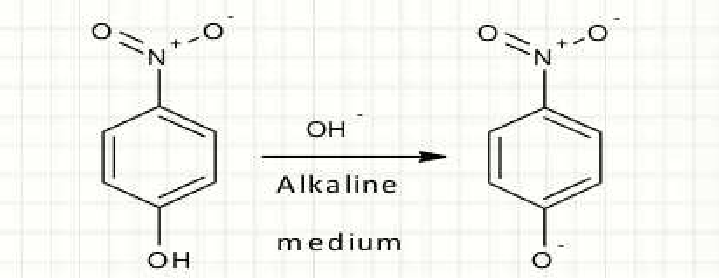
Blue Shift
(Hypsochromic)
Peaks shift to
shorter wavelength.

Red Shift
(Bathochromic)
Peaks shift to longer
wavelength.



Aniline

λ_{max} = 280 nm λ_{max} = 265 nm



p-nitrophenol

λ_{max} = 255 nm λ_{max} = 265 nm

Kolorimetrie

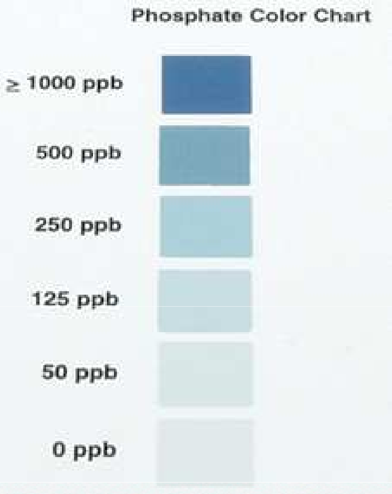
Fosfor

pH

Železo

Dusík (amoniakální, nitrátový)

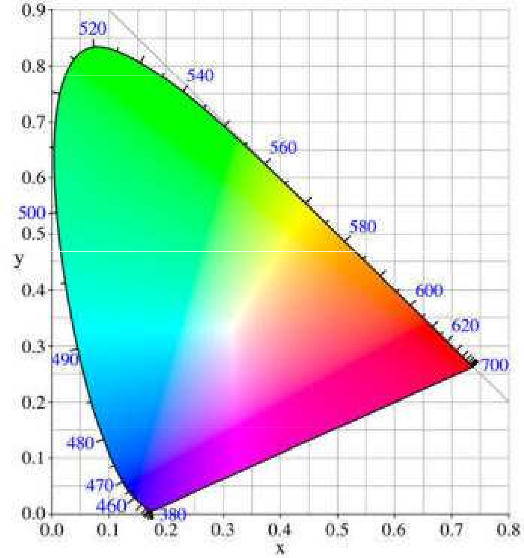
Kolorimetrie



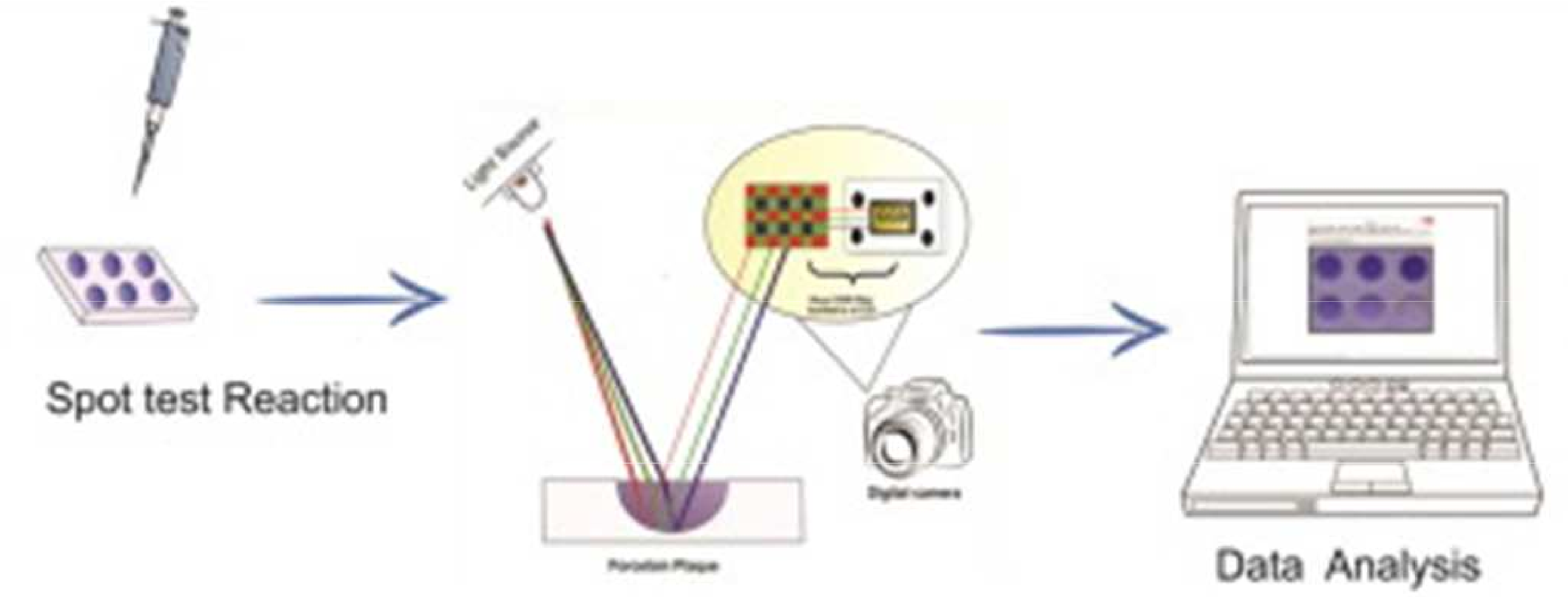
Phosphate Color Chart

≥ 1000 ppb	▨
500 ppb	▨
250 ppb	▨
125 ppb	▨
50 ppb	▨
0 ppb	▨

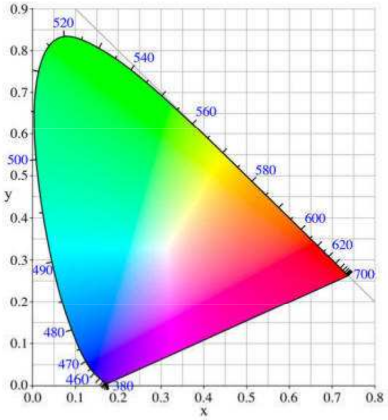
Signal processing



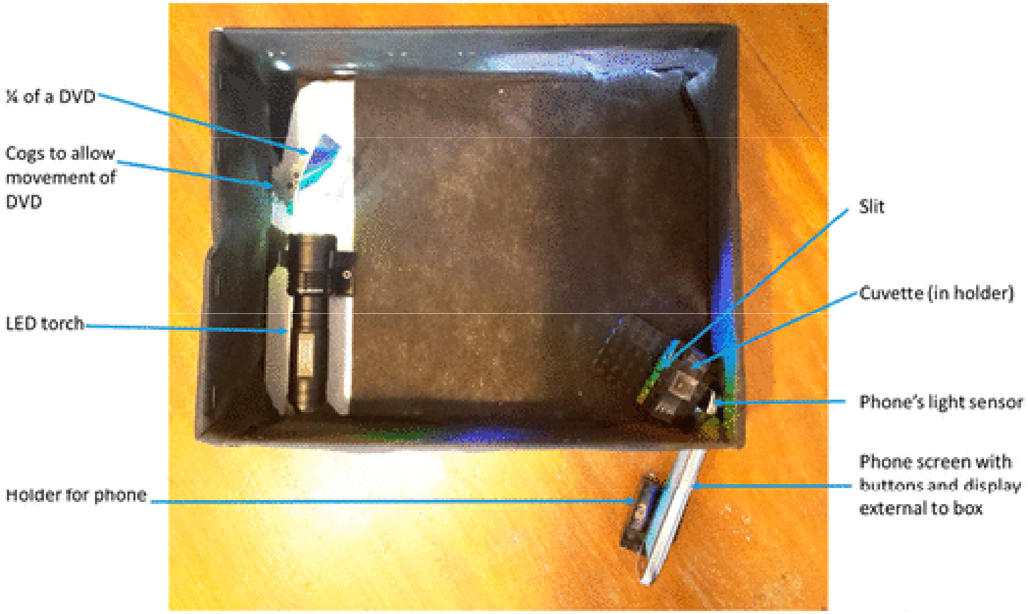
Kolorimetrie

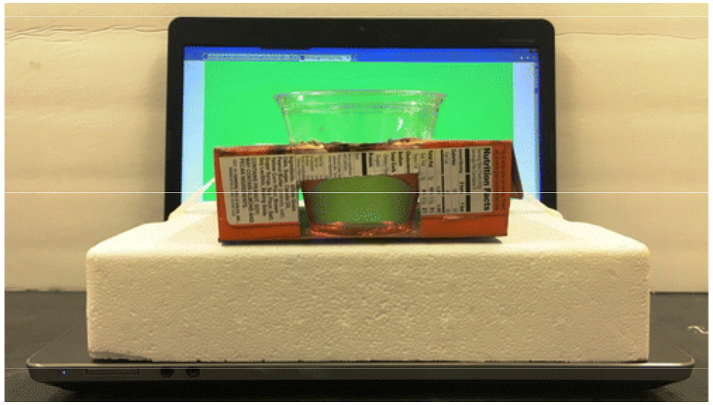


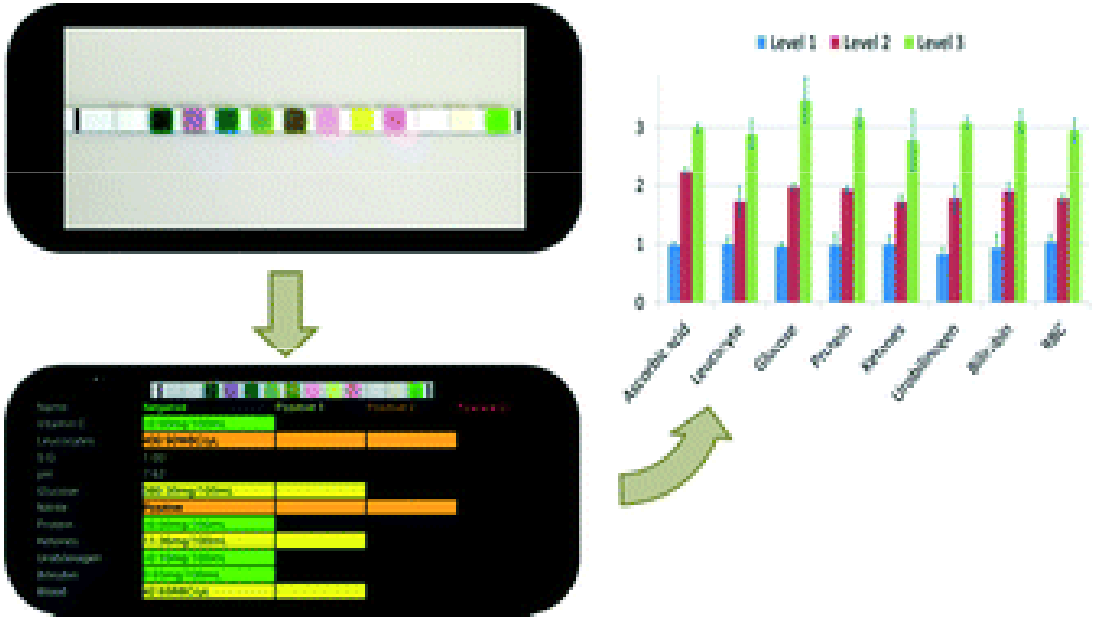
Signal processing



Kolorimetrie - aplikace mobilního telefonu







Atomová absorpční spektrometrie

Roztoková analýza

destruktivní

V plameni

V kyvetě

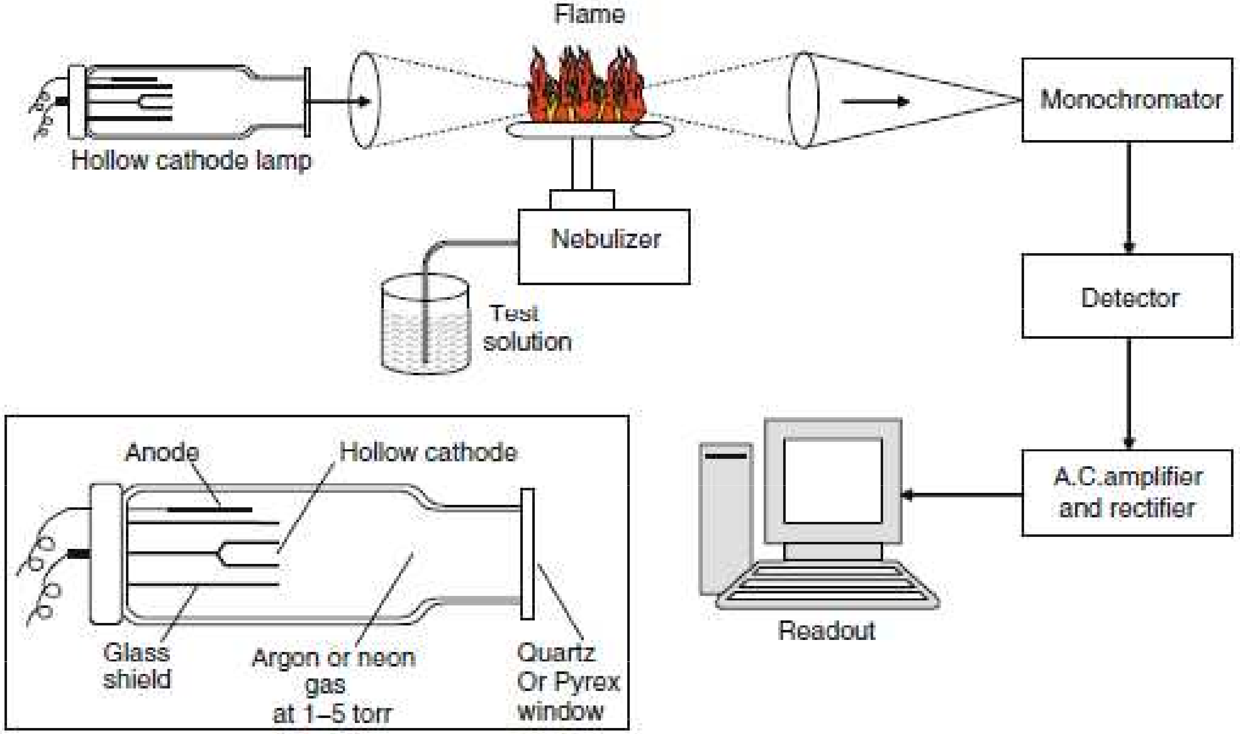
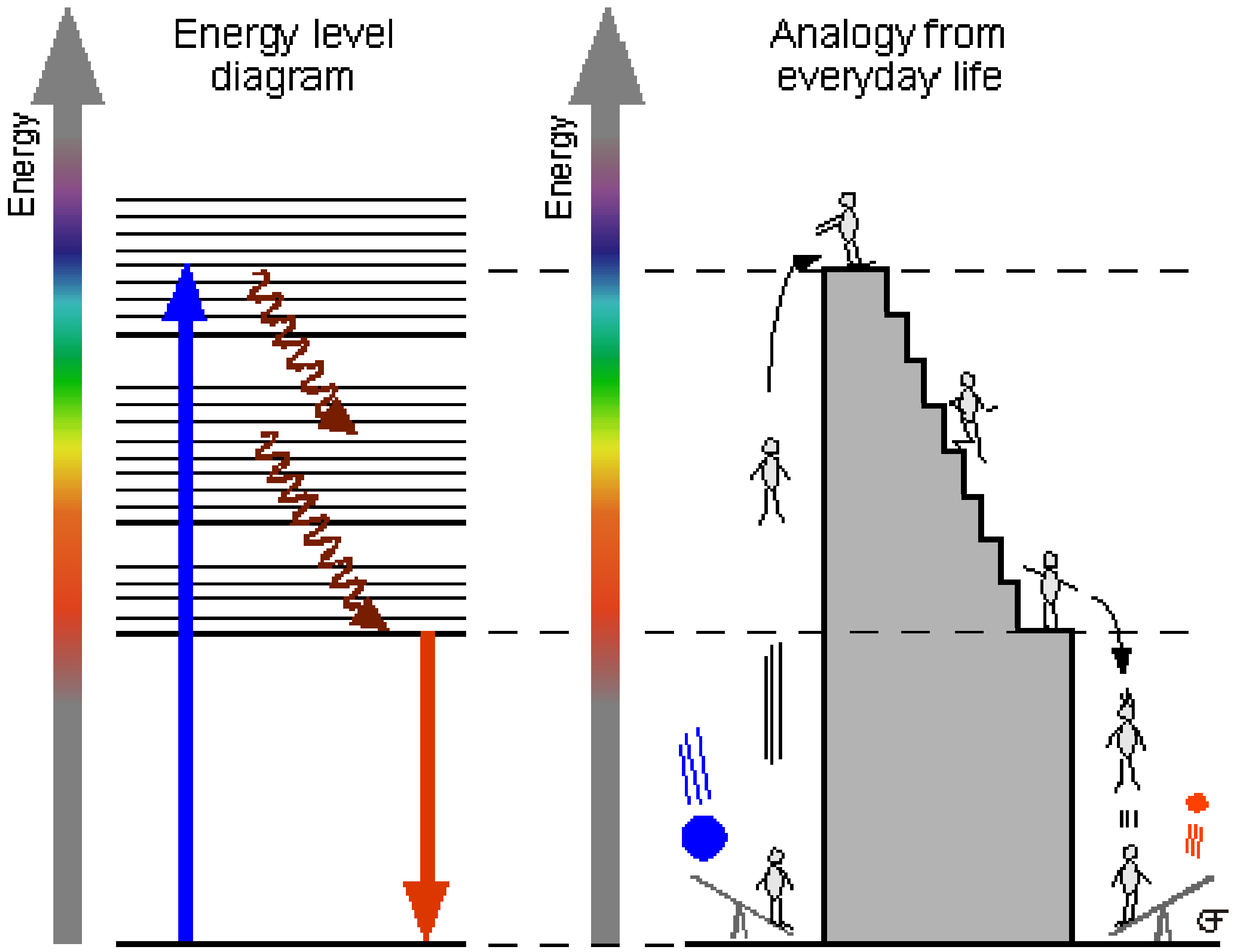


Fig. 34.1 Atomic absorption with hollow cathode lamp (From SKOOG. Principles of Instrumental Analysis, 5E. © 1998 Brooks/Cole, a part of Cengage Learning, Inc. Reproduced by permission. www.cengage.com/permissions)

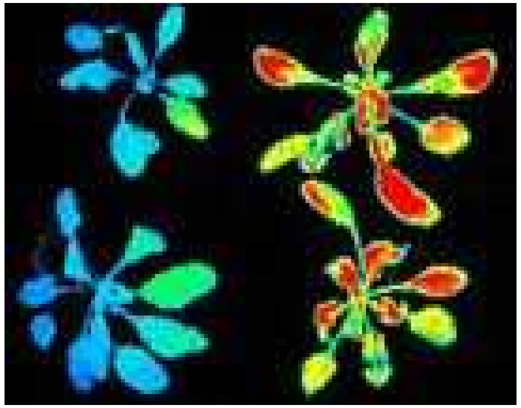
Fluorescence

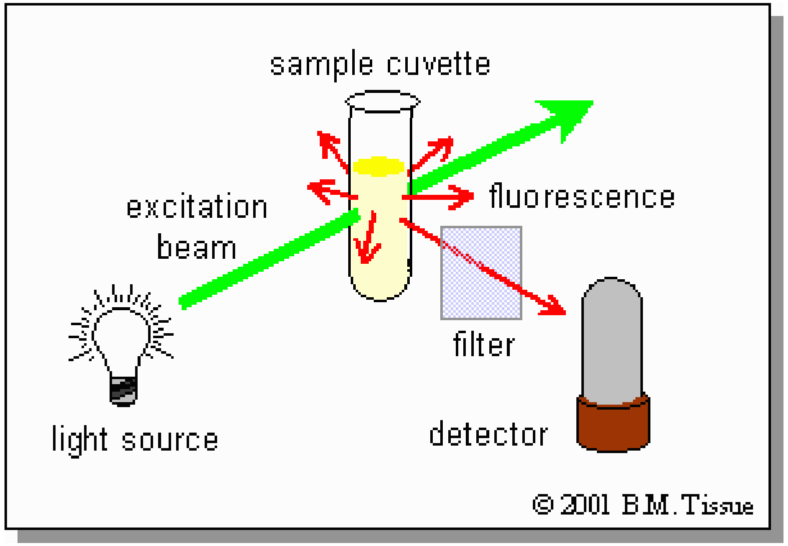


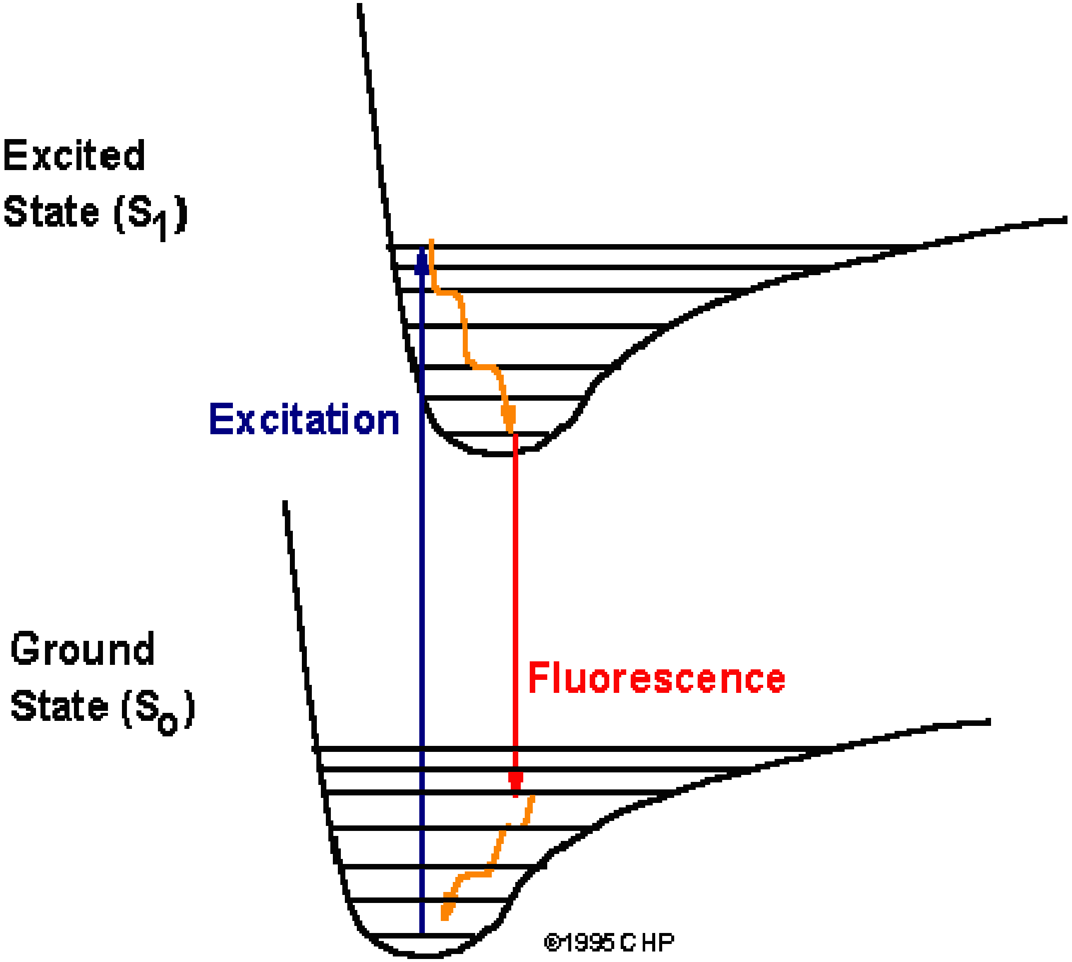
Absorption, Nonradiative Relaxation and Luminescence

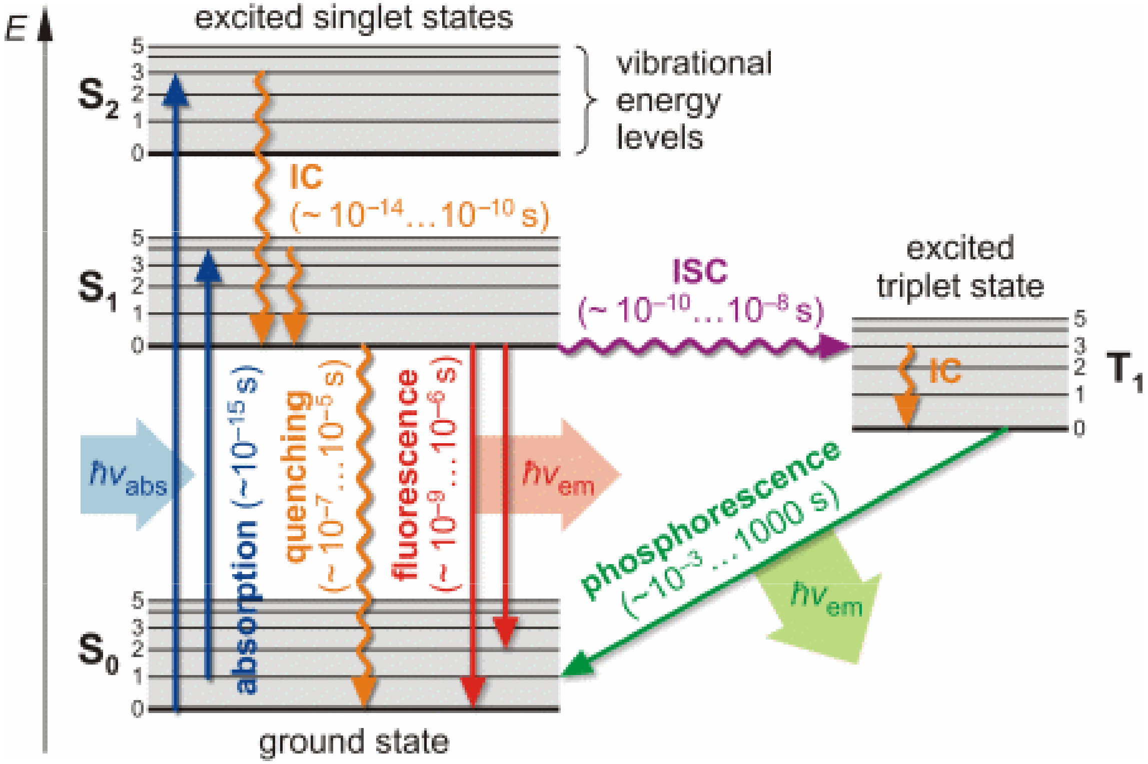
Making heat and 1x red out of 1x blue

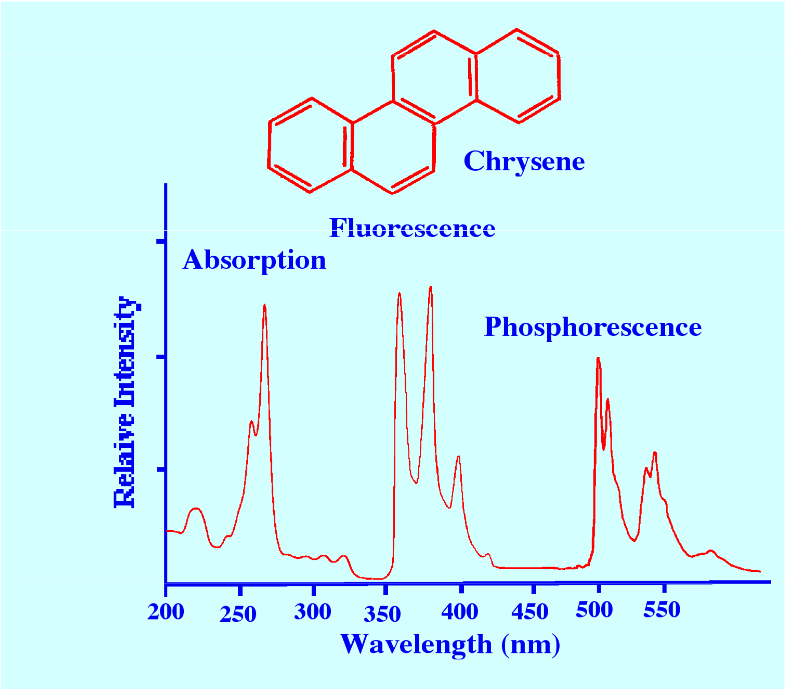
Fluorescence

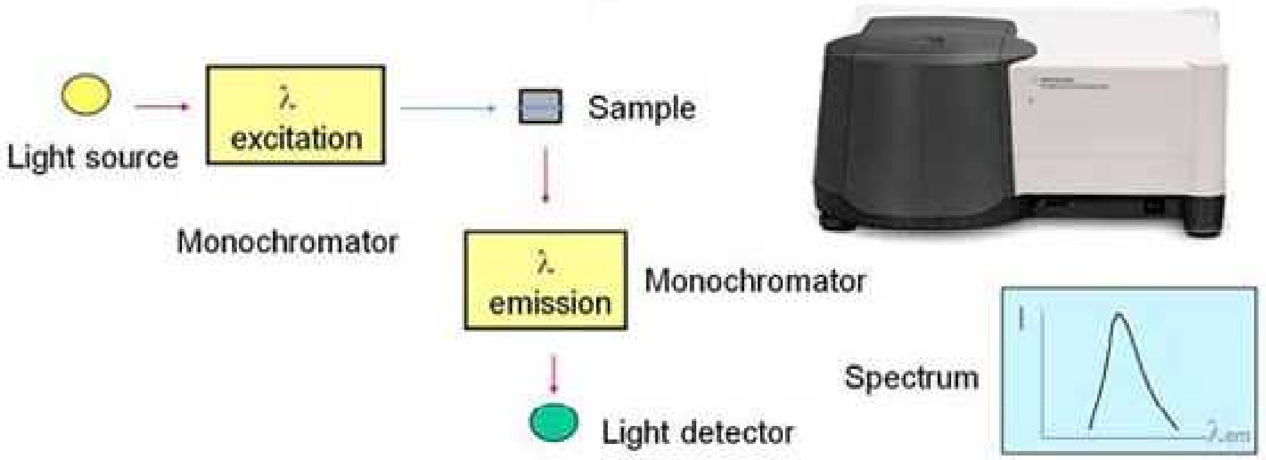






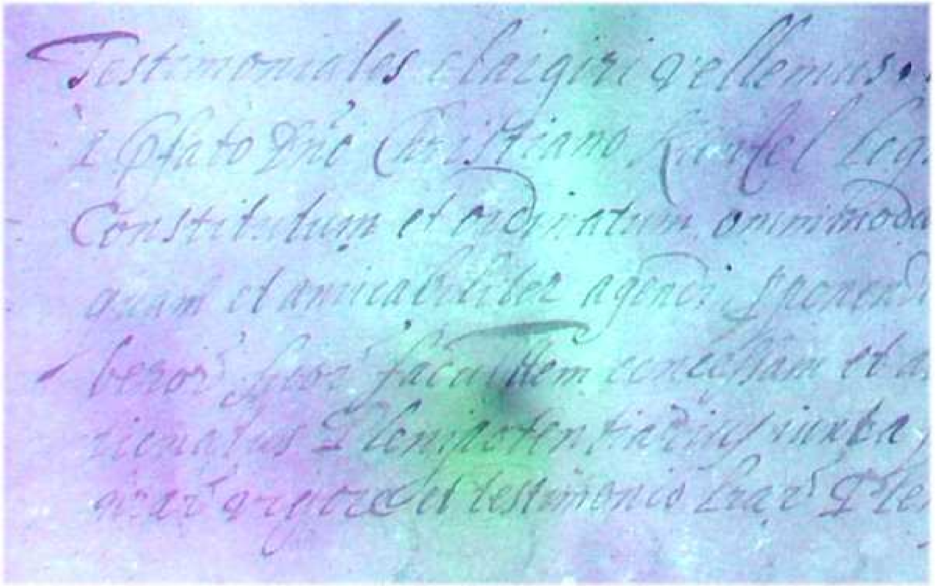

E
excited singlet states
vibrational energy levels
S2
S1
S0
5 3 2 1 0
IC
(~ 10−14…10−10 s)
ISC
(~ 10−10…10−8 s)
excited triplet state
T1
IC
ħνabs
absorption (~10−15 s)
quenching (~10−7…10−5 s)
fluorescence (~ 10−9…10−6 s)
ħνem
phosphorescence (~10−3 …1000 s)
ħνem
ground state


Chrysene
Fluorescence
Absorption
Phosphorescence
Relaive Intensity
200
250
300
350
400
450
500
550
Wavelength (nm)

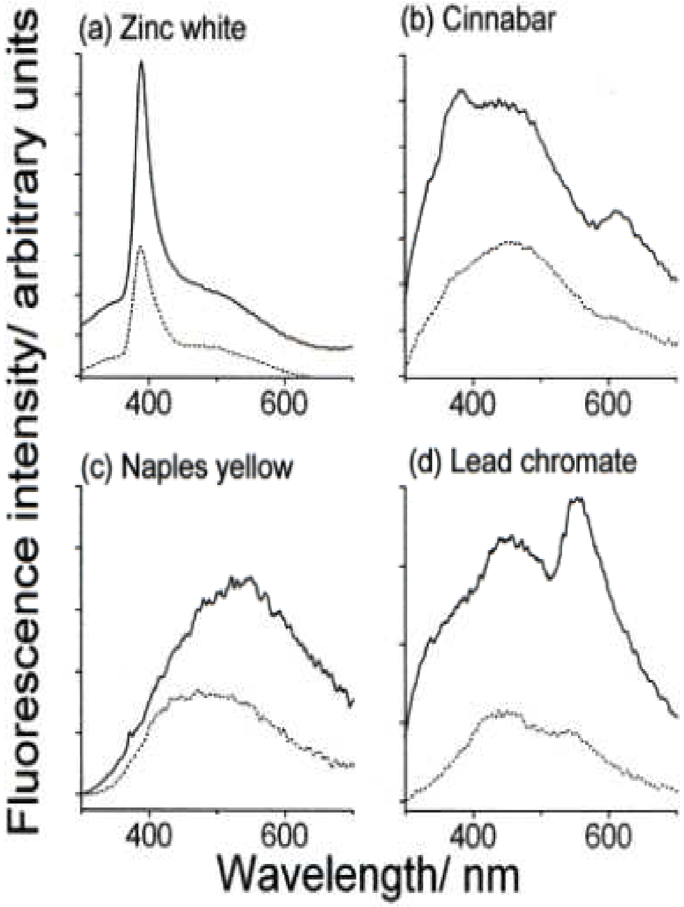

Light source
λ
excitation
Monochromator
Sample
λ
emission
Monochromator
Light detector
Spectrum
I
λem

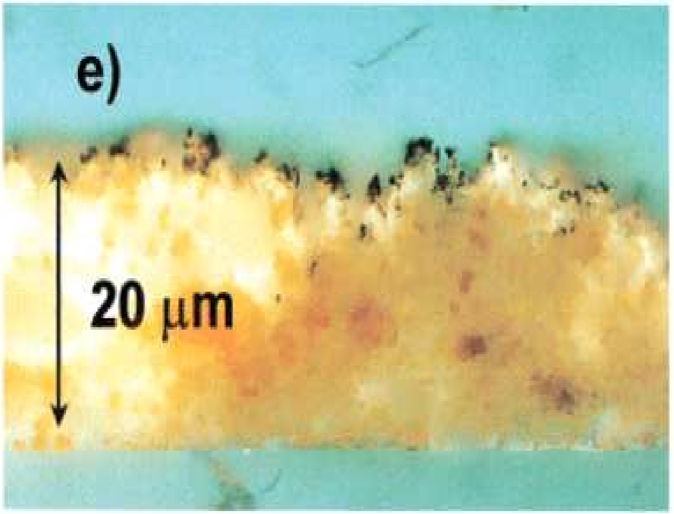
Fluorescence v UV světle

Aplikace ultrafialové fluorescence - zviditelnění časem degradovaného textu



Pigmenty





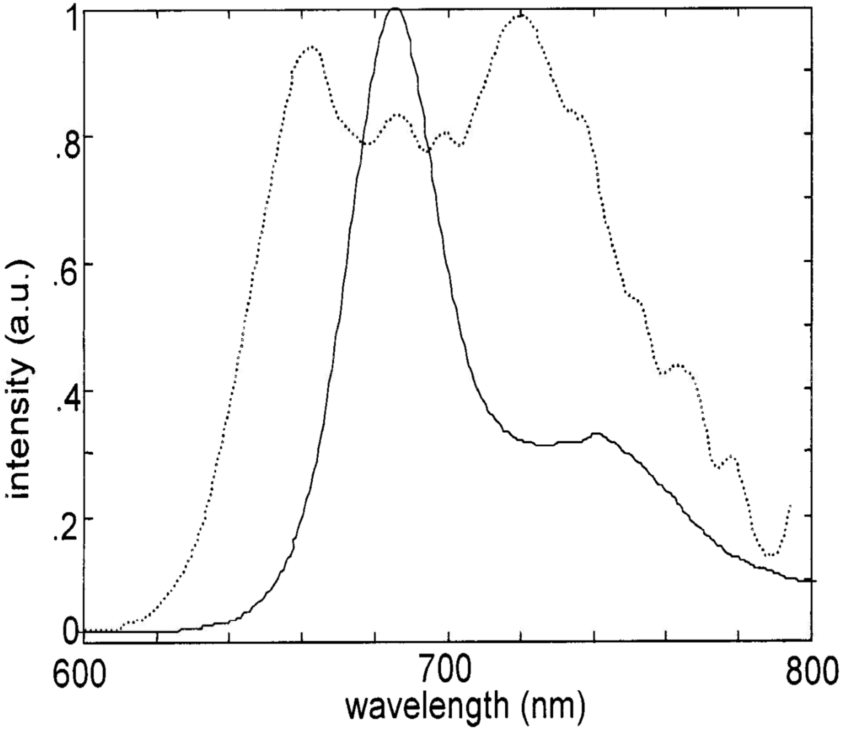
LIF spectra of unvarnished tempera systems taken at a resolution of 1 nm with a laser fluence of 1.2 mJ cm-2:

(a) zinc white,
(b) cinnabar,
(c) Naples yellow, and
(d) lead chromate.

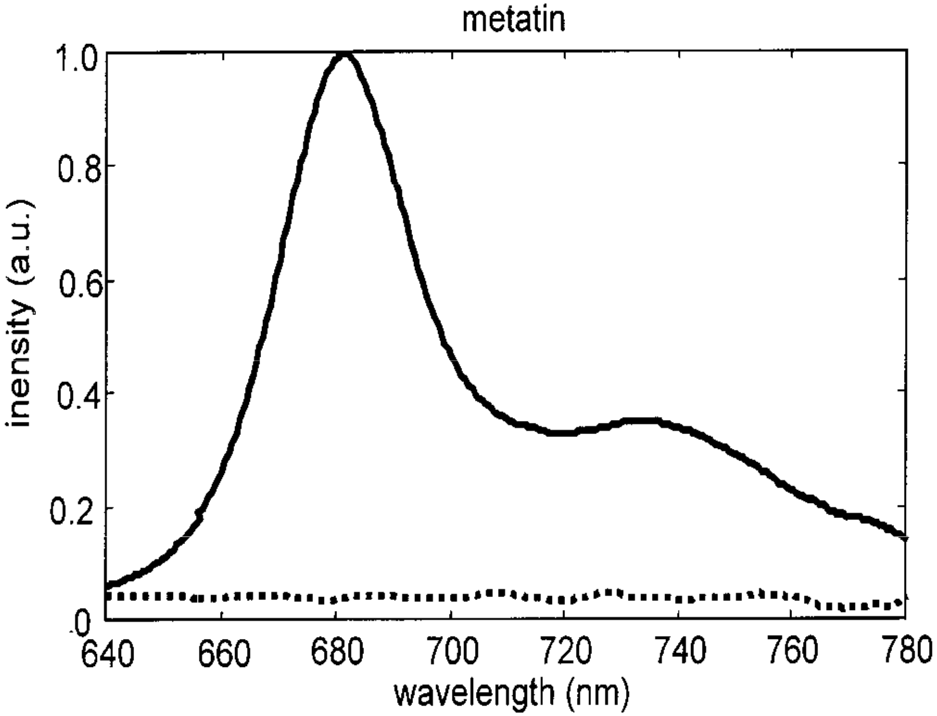
The exciting laser wavelength is 248 nm.

(e) UV fluorescence image of a cross section of an unvarnished laser-ablated region of a Naples yellow tempera sample. (Magnification is 500).

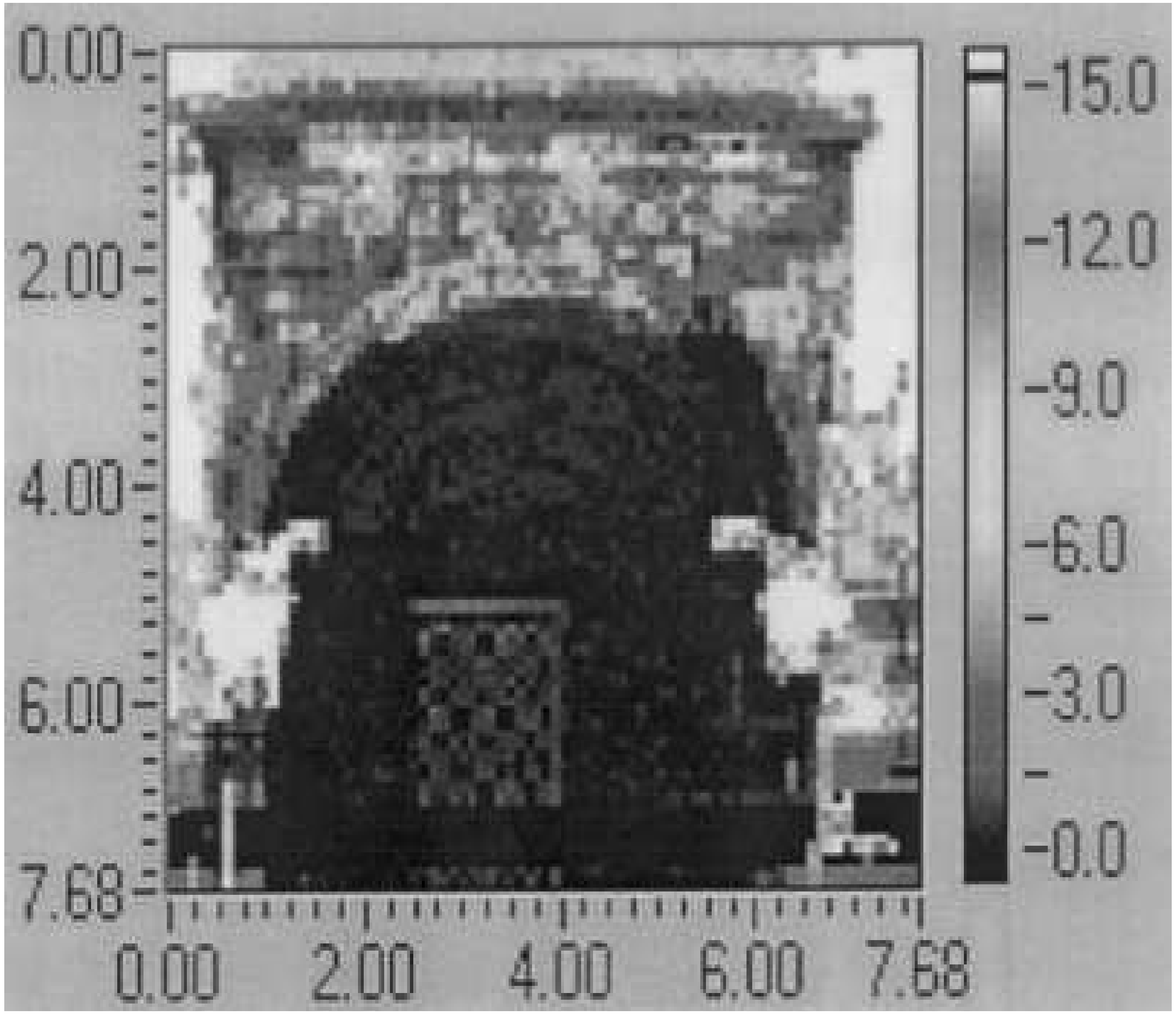
Detekce mikroorganismů



Normalised LIF spectra of green algae (continuous line) and cyanobacteria (dotted line). Excitation 355 nm.

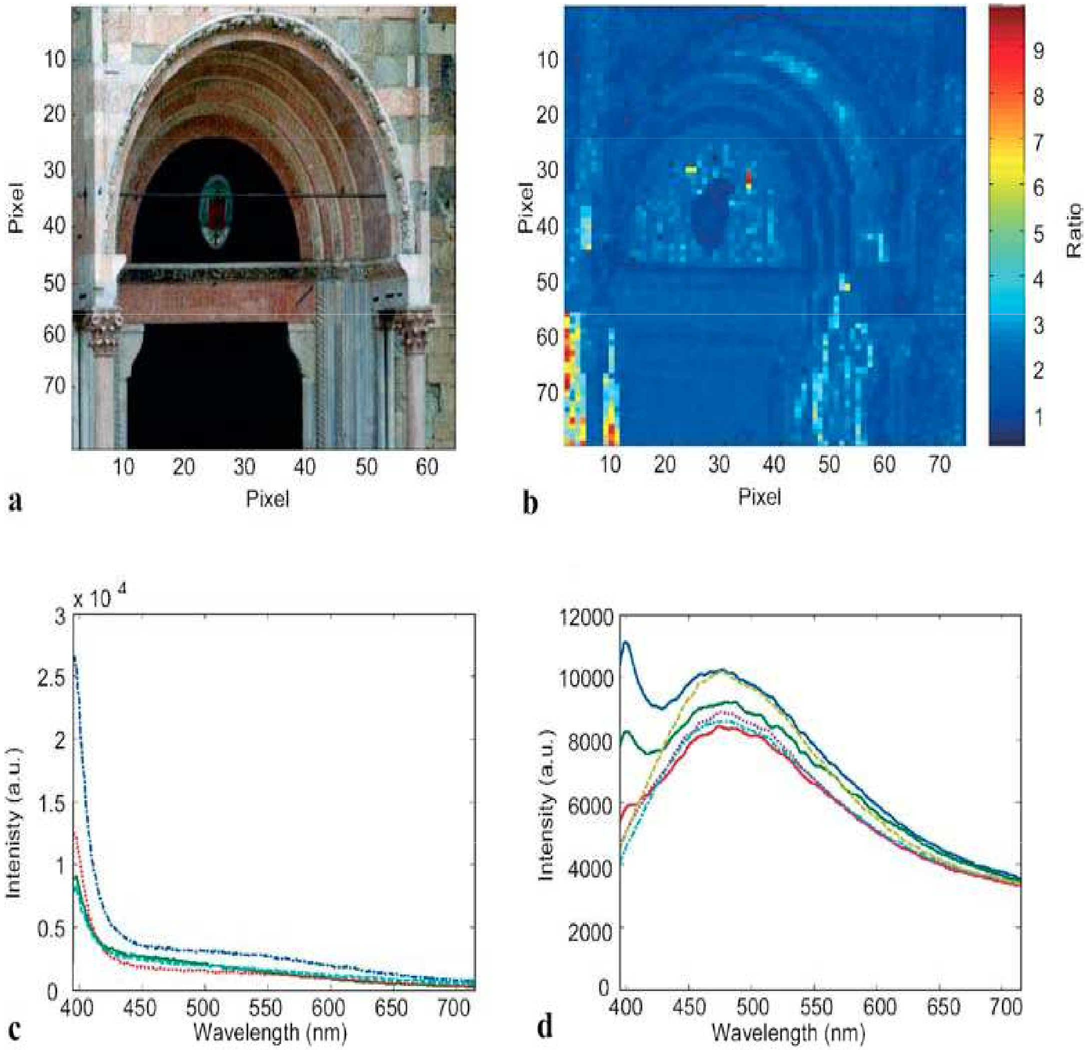


Fluorescence spectra of green algae on a marble substrate. Before a biocidal treatment (continuous line) and after (dotted line). Excitation wavelength 355 nm.



Fluorescence image related to the alga colonisation on the northern portal of **Lund Cathedral**. The intensity of the chlorophyll fluorescence in the band around 685 nm is indicated in grey levels and makes evident the important biodeteriogen colonisation on the stone surface.

Katedrála v Parmě



a a picture of the area investigated; **b** the thematic map obtained from the ratio between the integrated area in the range 396 to 408 nm and the integrated area in the range 409 to 450 nmn (the *yellow-red areas* in the image indicate areas subject to protective treatment); **c** fluorescence spectra taken from the bottom left area of the protiro and referring to those pixels of the thematic map in **b** where the protective treatment was strongly present (*yellow–red pixels*); and **d** fluorescence spectra taken in the bottom right area of the protiro where the protective treatment is present in a lower degree

Chemiluminiscence

= emise viditelného záření (luminiscence) v důsledku chemické reakce.

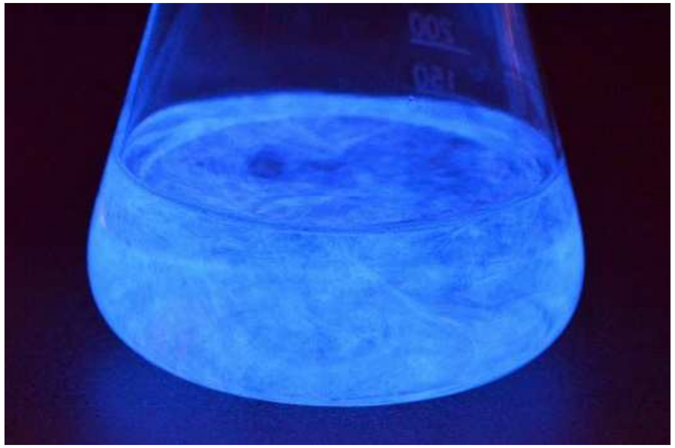
Např. luminol v přítomnosti peroxidu vodíku za přítomnosti vhodného katalyzátoru:

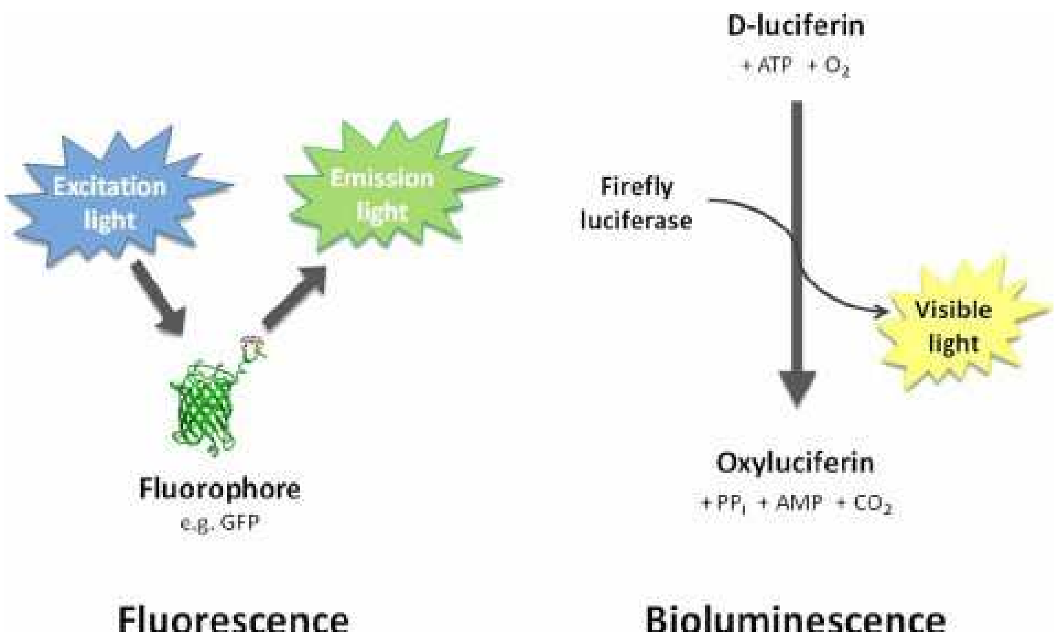
$$\underset{\text{luminol}}{C_8H_7N_3O_2} + \underset{\text{hydrogen peroxide}}{H_2O_2} \longrightarrow 3-\mathrm{APA}[\lozenge] \longrightarrow 3-\mathrm{APA} + \text{light}$$

kde:

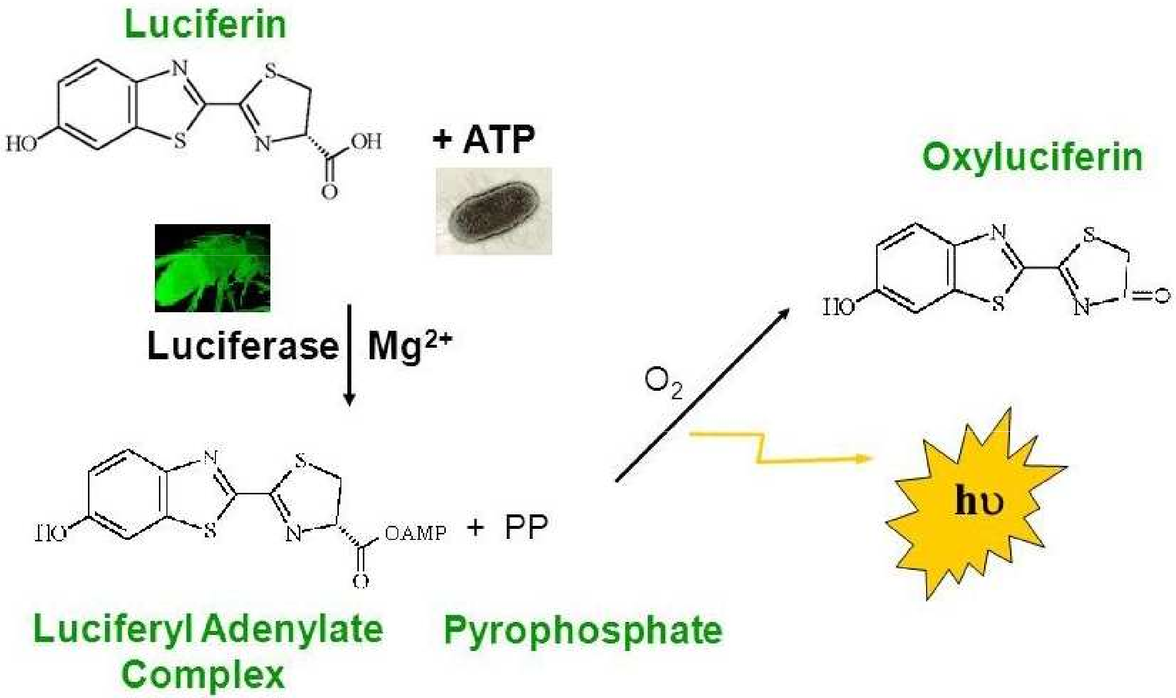
•3-APA je 3-aminophthalate

•3-APA[◊] is the vibronic excited state fluorescing as it decays to a lower energy level.





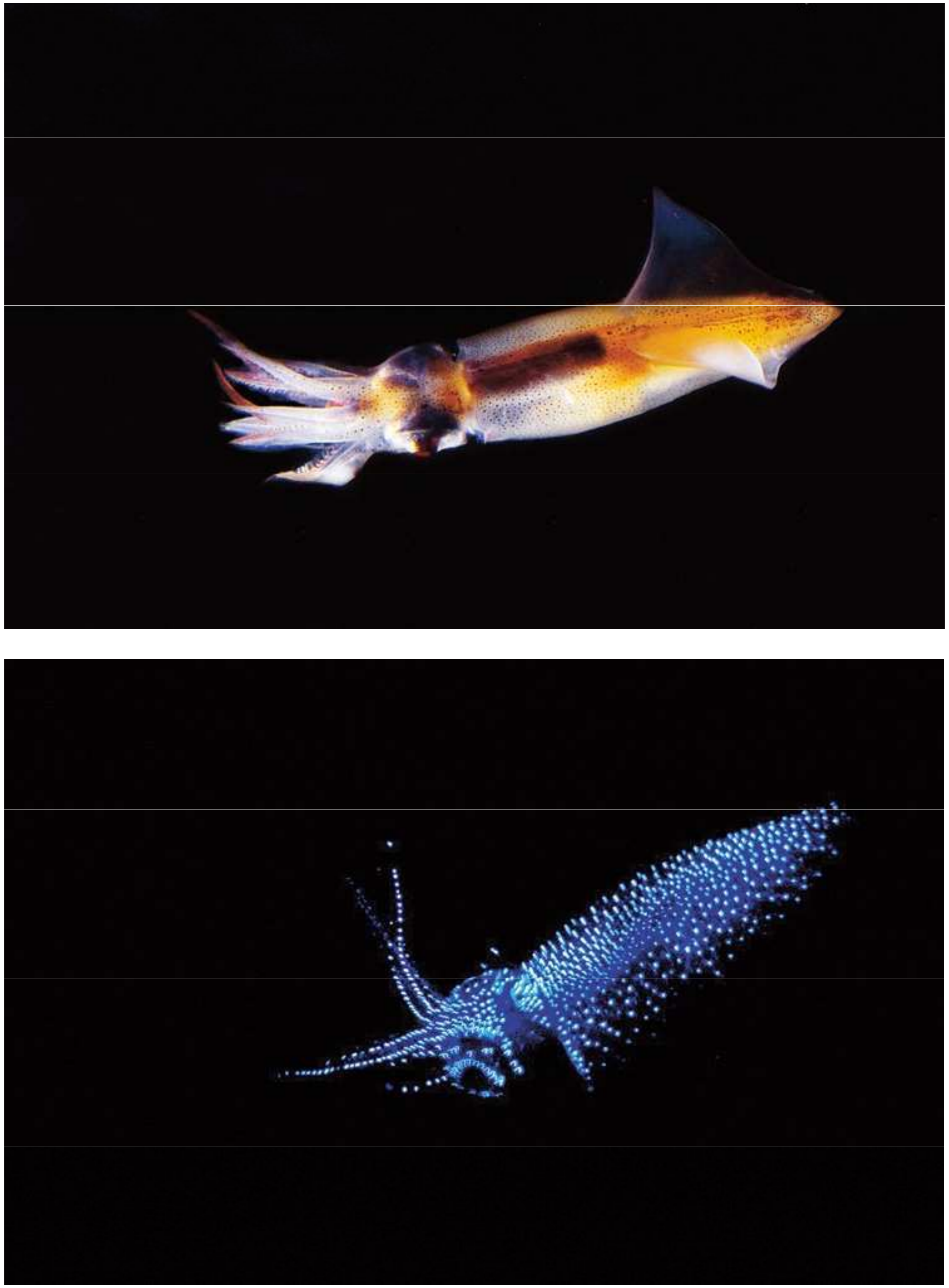
Lampyris noctiluca (světluška větší)



Pyrophorus noctilucus
(kovařík cucujo)

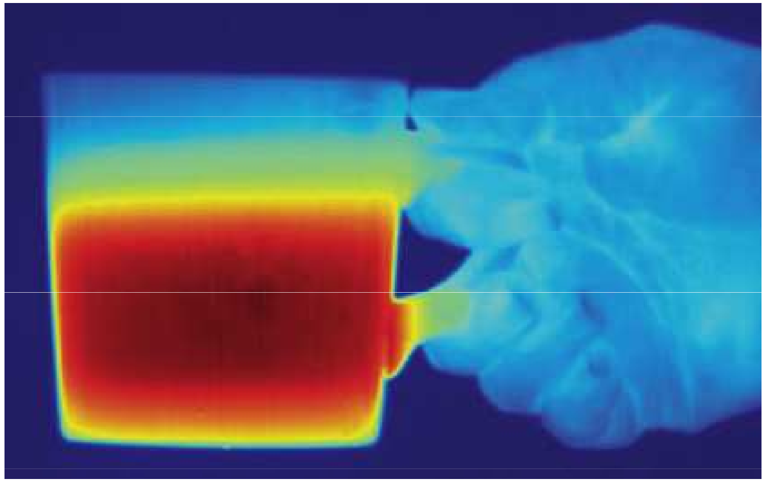
Svítivník malozubý (Pachystomias microdon)
svítí červeným světlem

Kalmar galérový
(Cranchia scabra

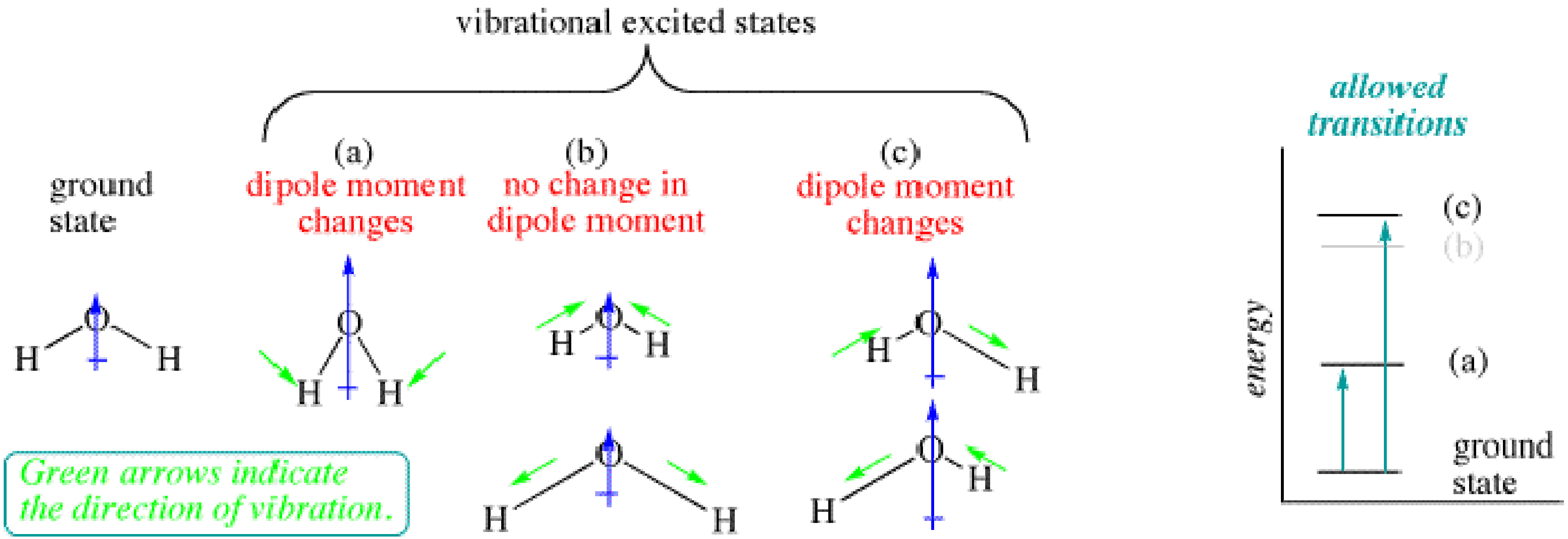


Hlubinná krakatice Watasenia scintillans v normálním
světle a "rozsvícená" v temné hlubině

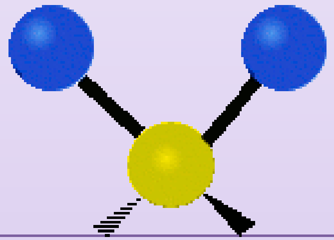
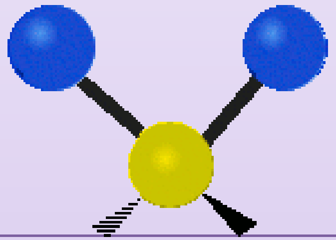
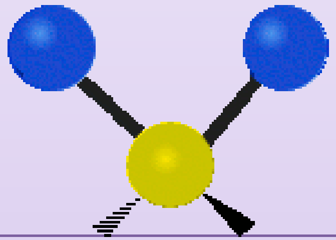
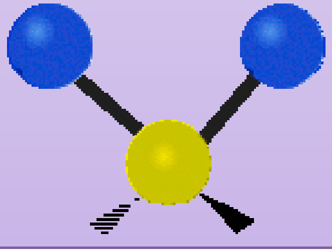
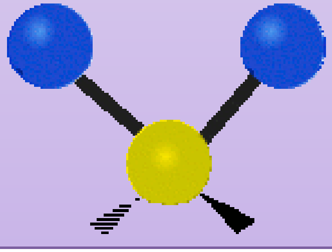
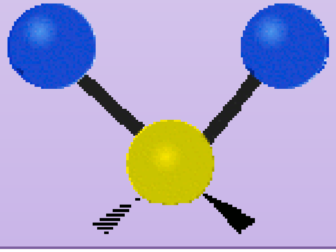
Infračervené záření (IR)



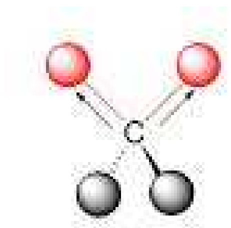
= tepelné záření, neviditelné okem, jeho zdrojem je každé těleso které má teplotu vyšší než je absolutní nula. Při pohlcování se IR záření mění na vnitřní energii pohlcujícího tělesa (těleso se ohřívá).



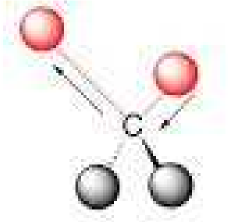
Vibrations of Molecules

Symmetrical stretching	Antisymmetrical stretching	Scissoring
		
Rocking	Wagging	Twisting
		

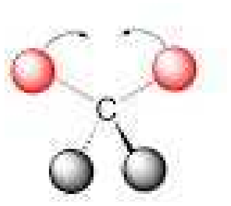
Infračervená spektrometrie (FTIR)



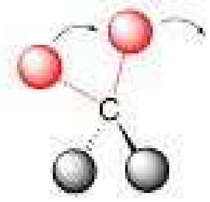
symmetric stretching



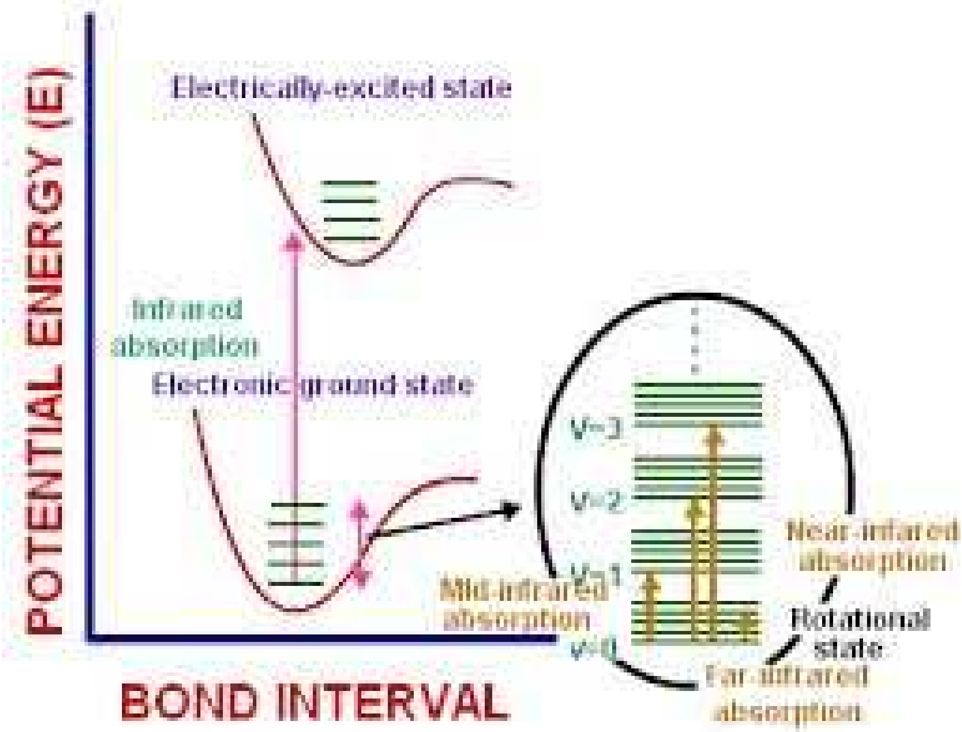
asymmetric stretching

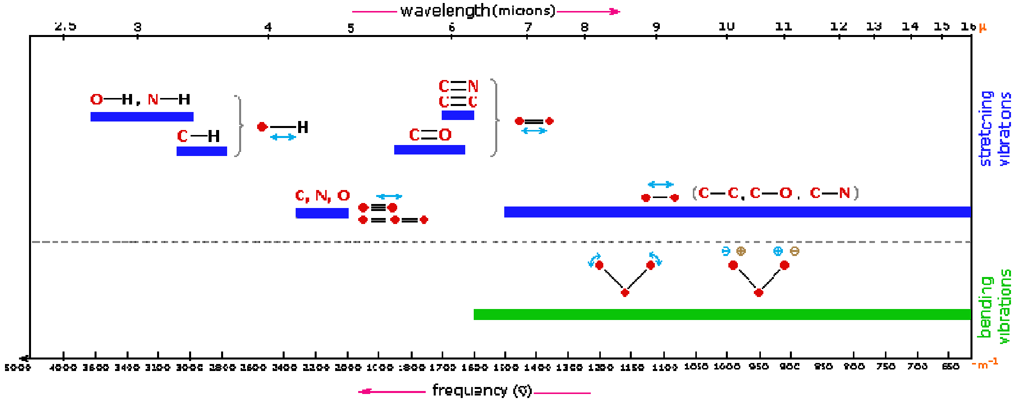


scissoring



rocking

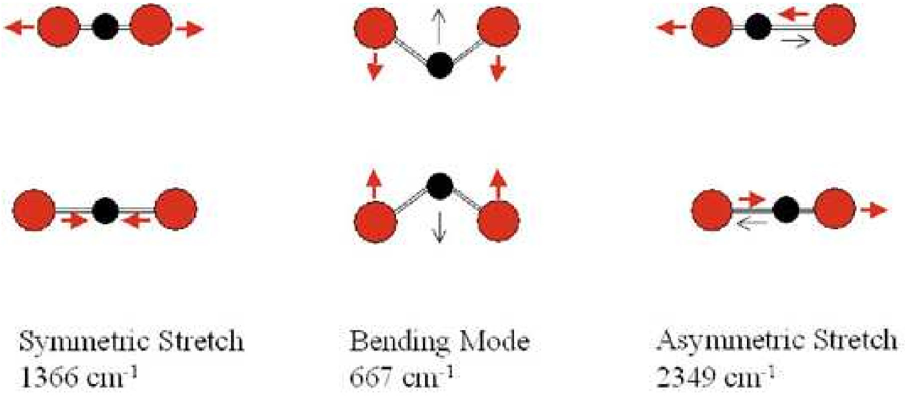


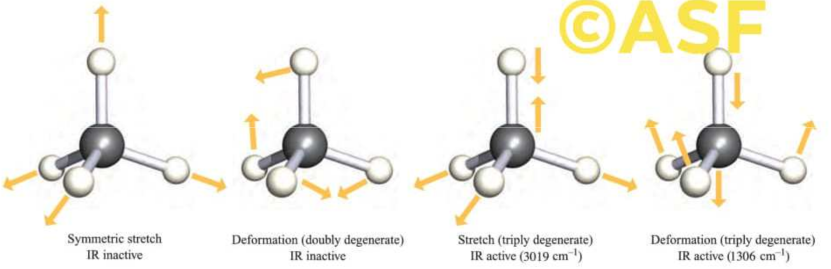


Příklad

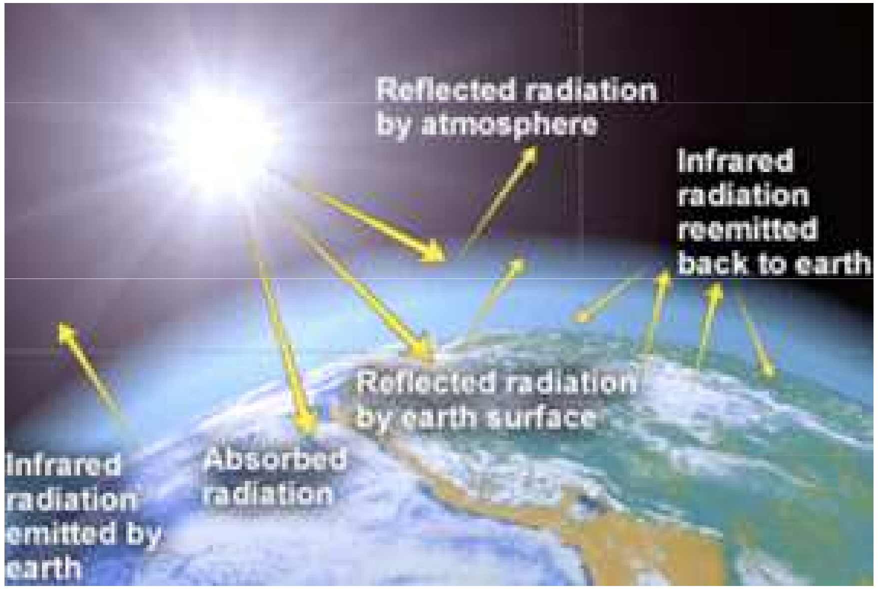
Oxid uhličitý nemá v základním stavu molekulový dipól. Některé vibrace molekul CO_2 vytvářejí struktury s molekulovým dipólem. Z tohoto důvodu CO_2 silně absorbuje infračervené záření. Podobně se chová i *methan, vodní pára* a další **skleníkové plyny**. Tyto plyny absorbují infračervené záření ze zemského povrchu a emitovat je zpět ve všech směrech.

U hlavních plynů atmosféry, N_2 a O_2, k absorpci nedochází a záření prochází atmosférou do vesmíru.

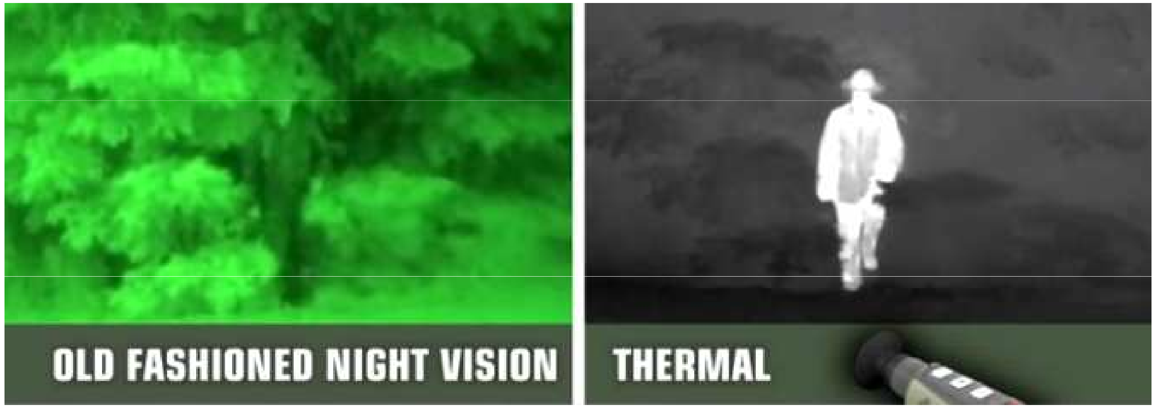




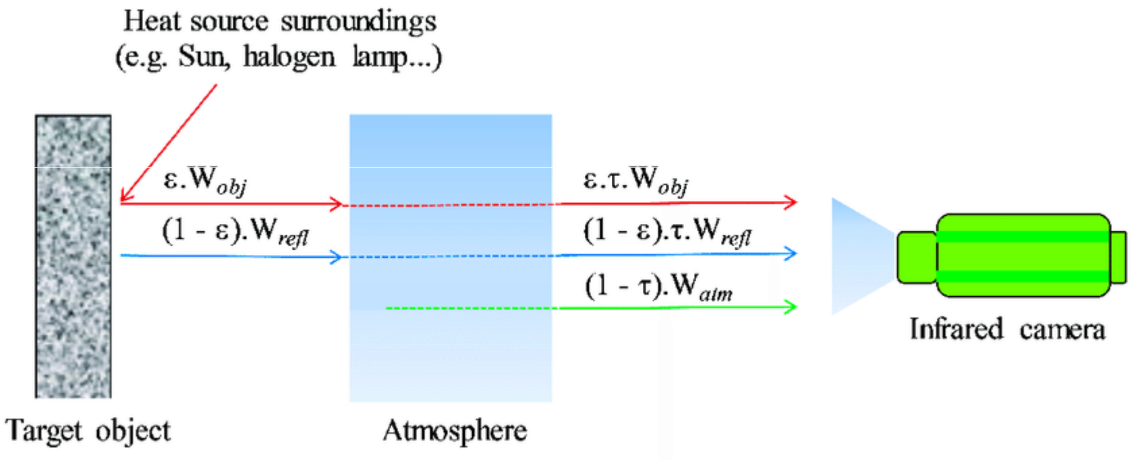
Gas	Current concentration	Residence time, in years	Relative global warming efficiency, 100-year horizon
CO_2	373 ppm	50–200	1
CH_4	1.77 ppm	12	23
N_2O	316 ppb	120	296
CFC-11	0.26 ppb	45	4600
HCFC-22	0.15 ppb	12	1700
HFC-134a	0.01 ppb	14	1300
Halon-1301	0.003 ppb	65	6900



Noční vidění a termokamery

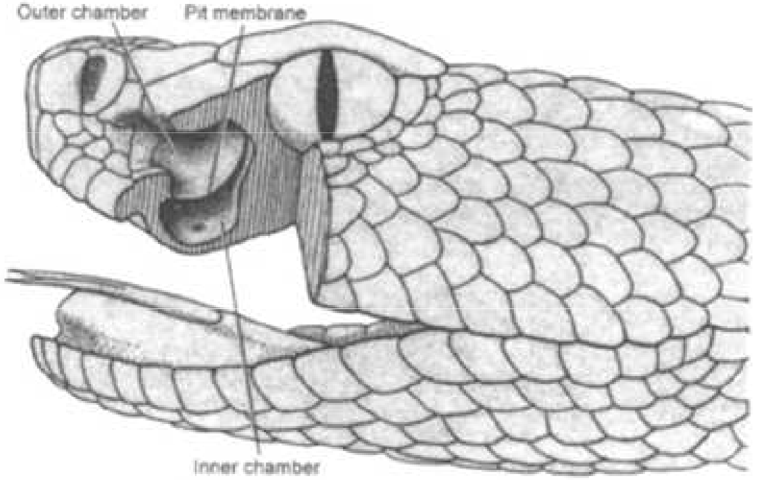


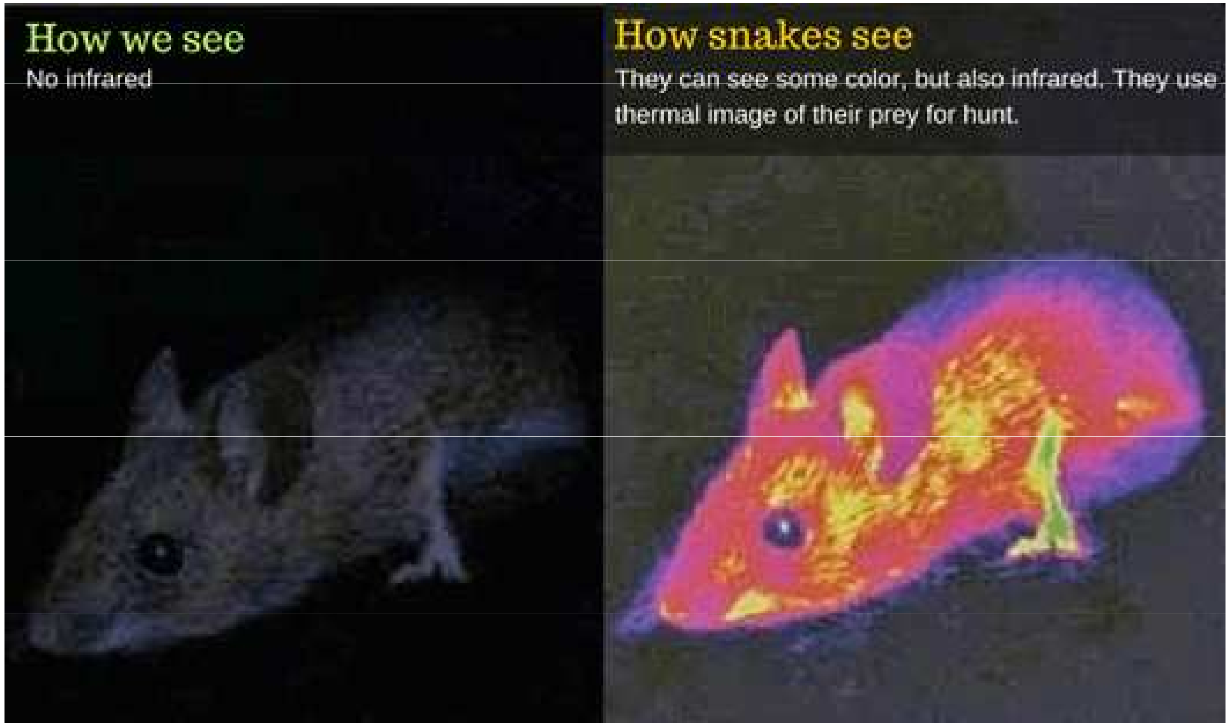
Termografie = analýza infračervené energie vyzařované tělesem. Termografickým měřicím systémem lze zobrazit teplotní pole na povrchu sledovaného objektu. Infračervené záření je pro lidské oko neviditelné, proto se termovizní snímky vizualizují za použití okem viditelných palet, které přiřazují barvu různým teplotám (různému množství tepelného toku).



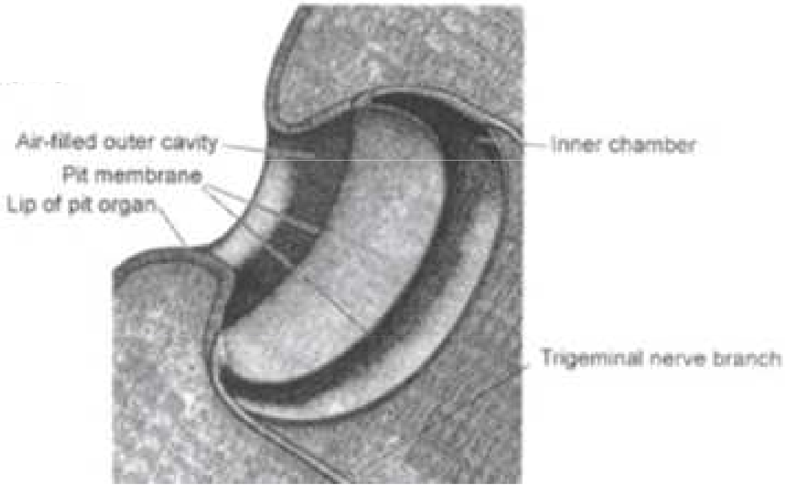
Detekce infračerveného záření u hadů

Zmije, krajty a hroznýši mají na tváři otvory, tzv. <u>jamkové orgány</u>, které obsahují membránu schopnou detekovat infračervené záření až do vzdálenosti jednoho metru. V noci umožňují hadům "vidět" obraz predátora nebo kořisti (podobně jako infračervená kamera).



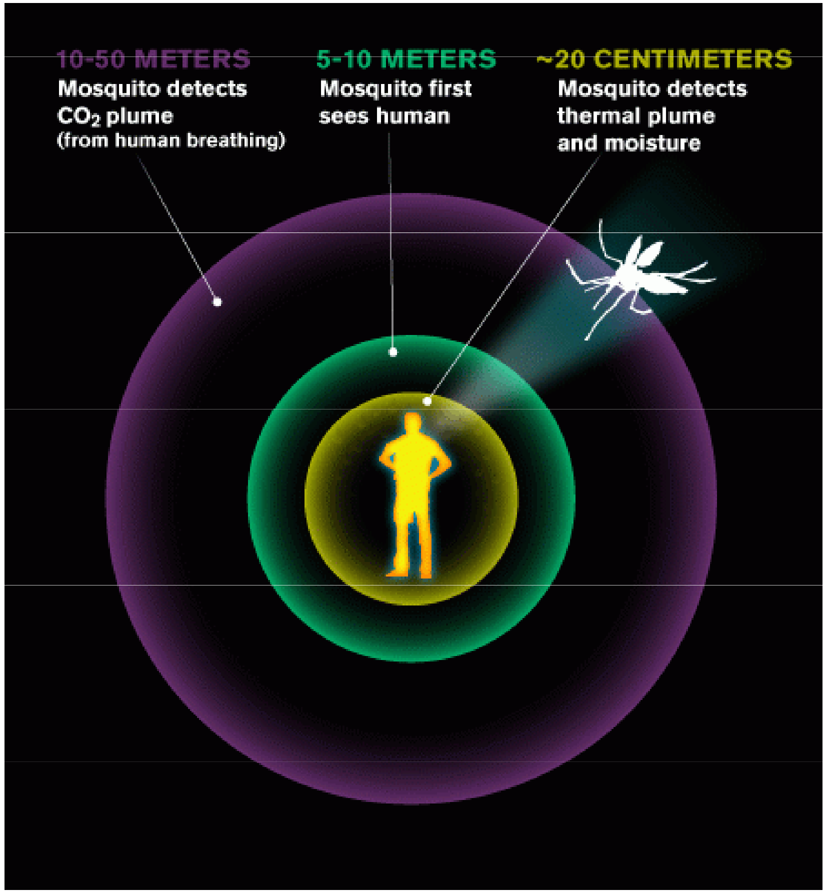


They can see some color, but also infrared. They use thermal image of their prey for hunt.

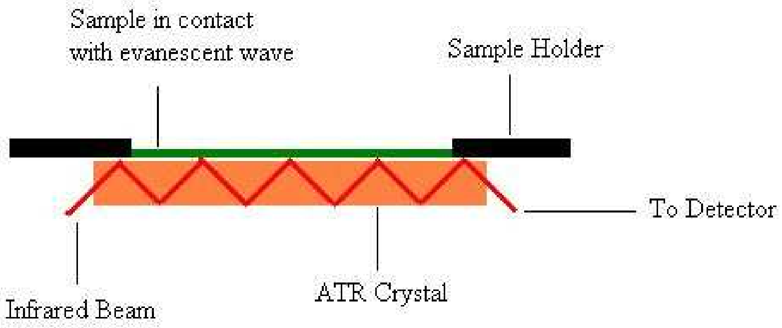


Upír obecný *(Desmodus rotundus)*

- čidlo tepelného záření se nachází v čumáku upíra, anatomicky je podobné obdobnému orgánu hadů.

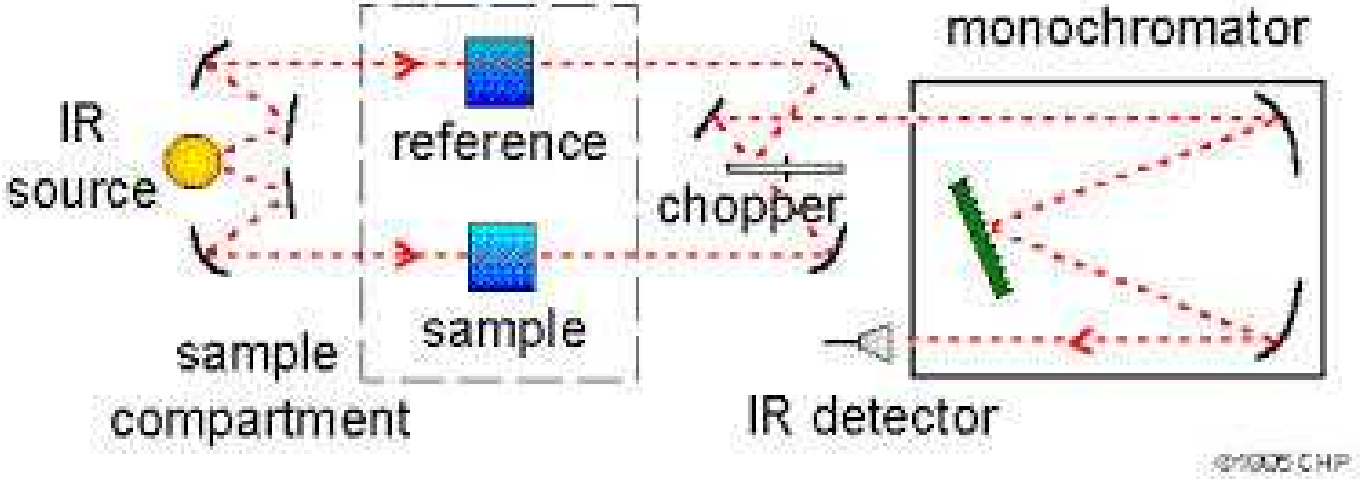


Infračervená spektrometrie



Schematic of a multiple reflection ATR system

ATR
KBr technika



Sample Preparation

- Gaseous samples

Long pathlength to compensate for the diluteness

- Liquid samples

Can be sandwiched between two plates of a salt

  - sodium chloride
  - potassium bromide
  - calcium fluoride

- Solids
  - KBr pellet
  - Nujol mull
  - Dissolving in organic solvent(CCl_4)

Infračervená spektrometrie s Fourierovou transformací (FTIR)

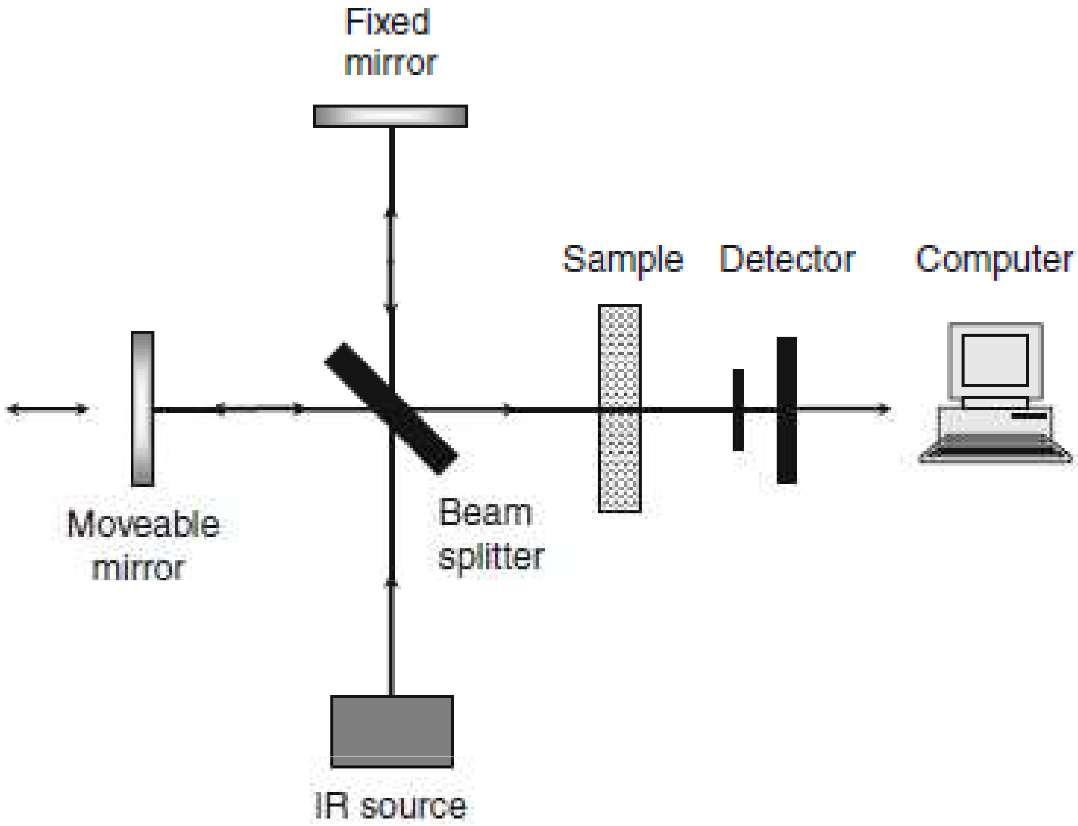
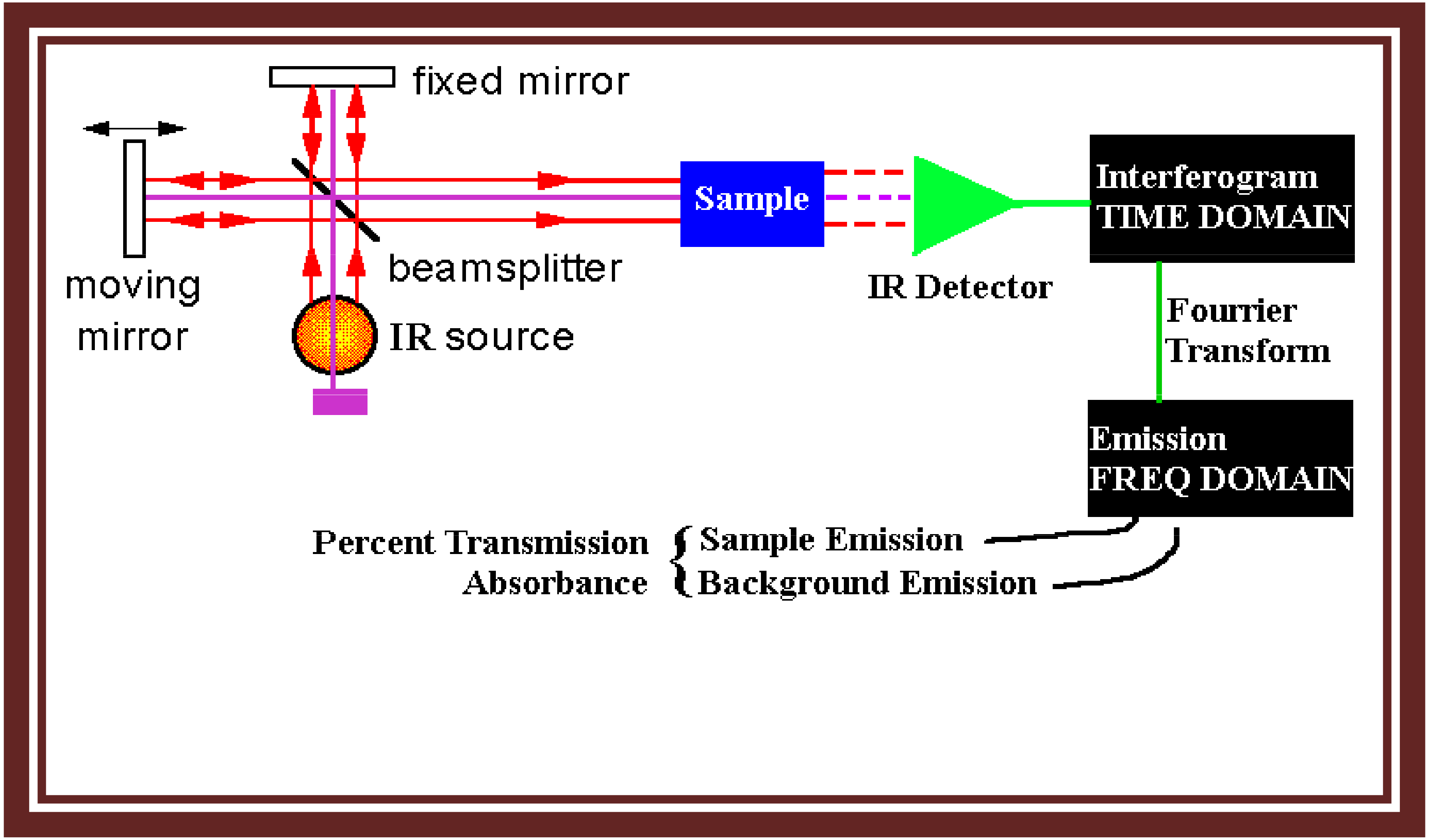
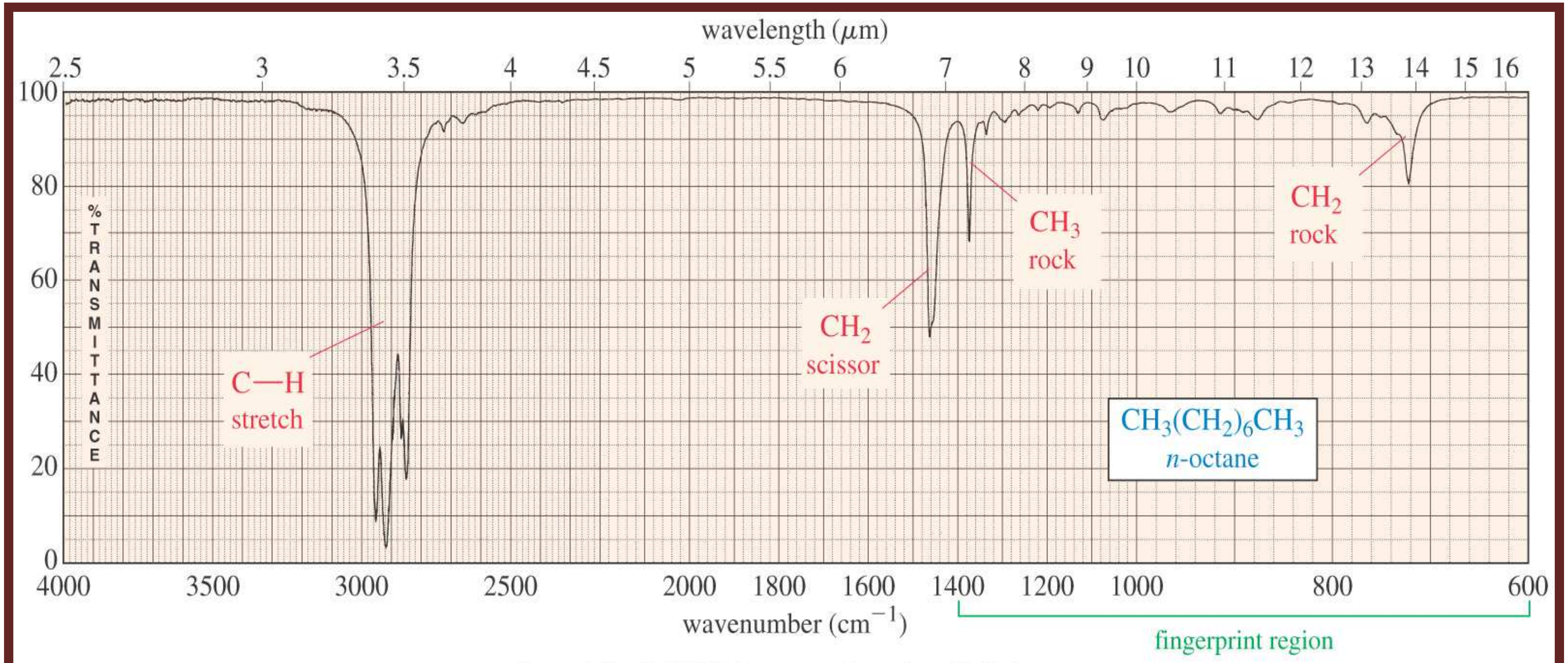


Fig. 35.6 FT-IR spectrometer with interferometer (Reprinted from "Applied Spectroscopy", Jerry Workman, Jr., page 9, Copyright 1998, with permission of Elsevier)

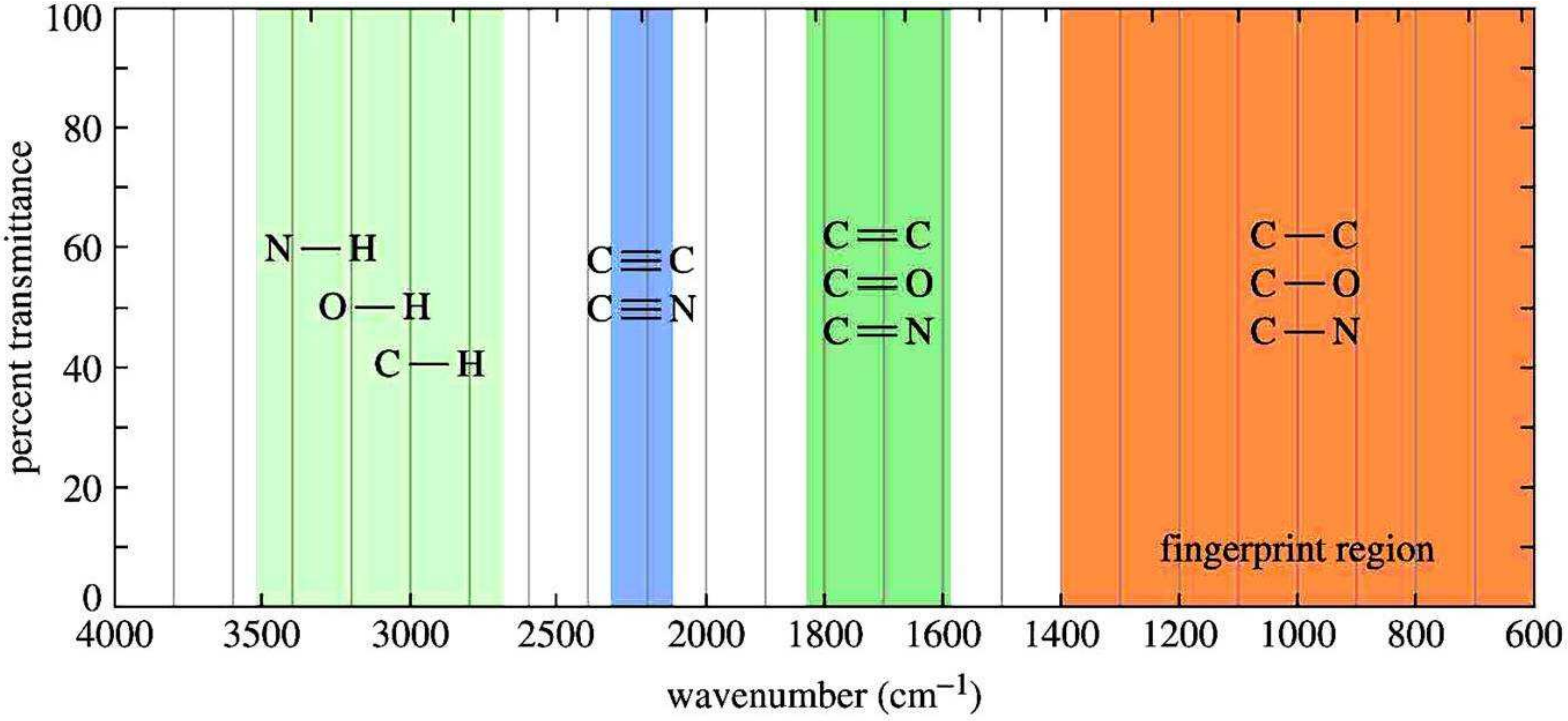
Infračervená spektrometrie s Fourierovou transformací (FTIR)



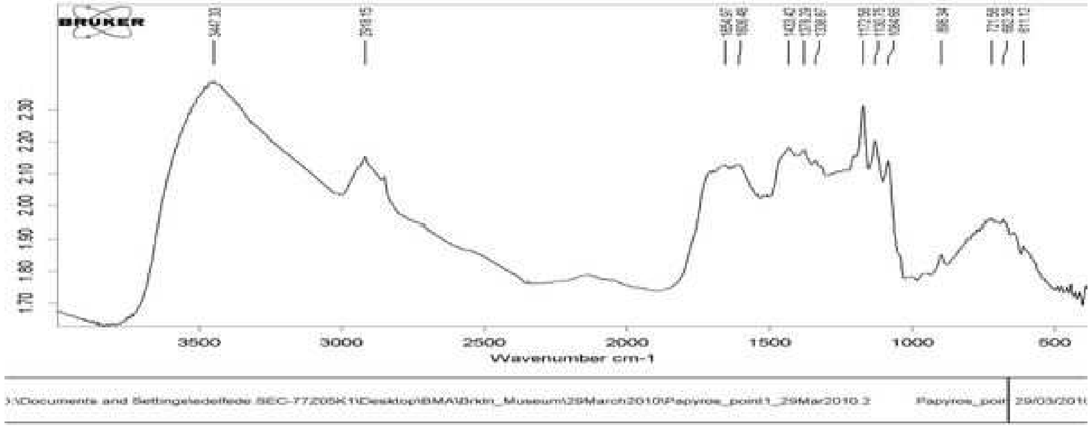
A Fourier transform converts the time domain to the frequency domain with absorption as a function of frequency.

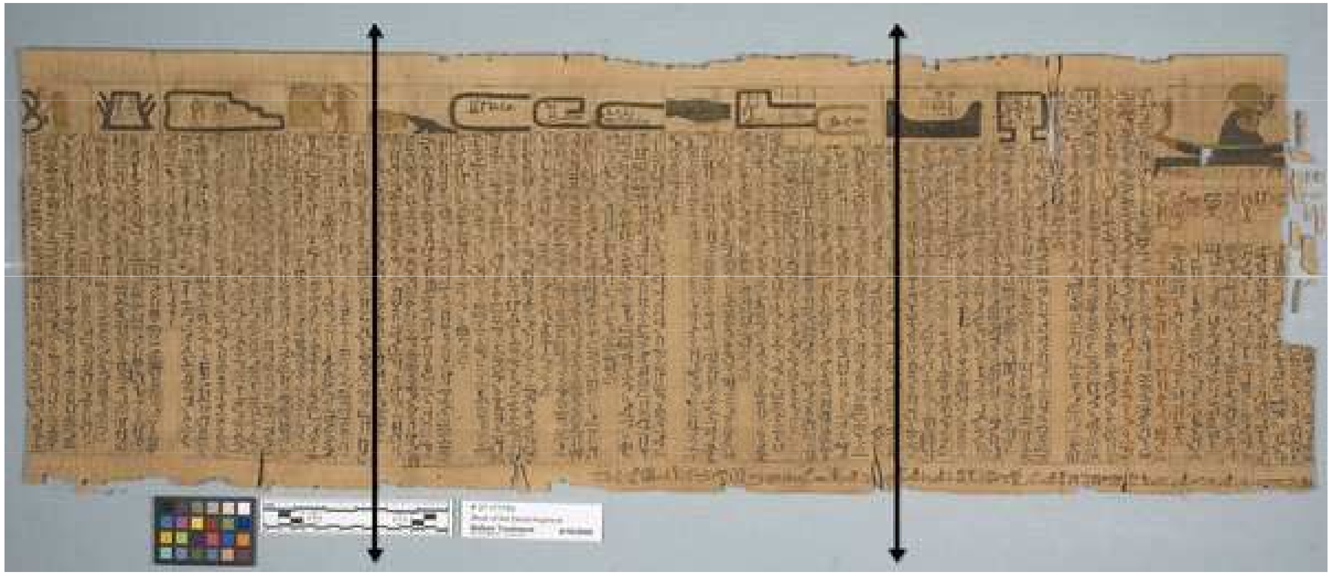


The typical IR absorption range for covalent bonds is **600 - 4000 cm^{-1}**. The graph shows the regions of the spectrum where the following types of bonds normally absorb. For example a sharp band around 2200-2400 cm^{-1} would indicate the possible presence of a C-N or a C-C triple bond.



Egyptská kniha mrtvých





Přenosné FTIR zařízení

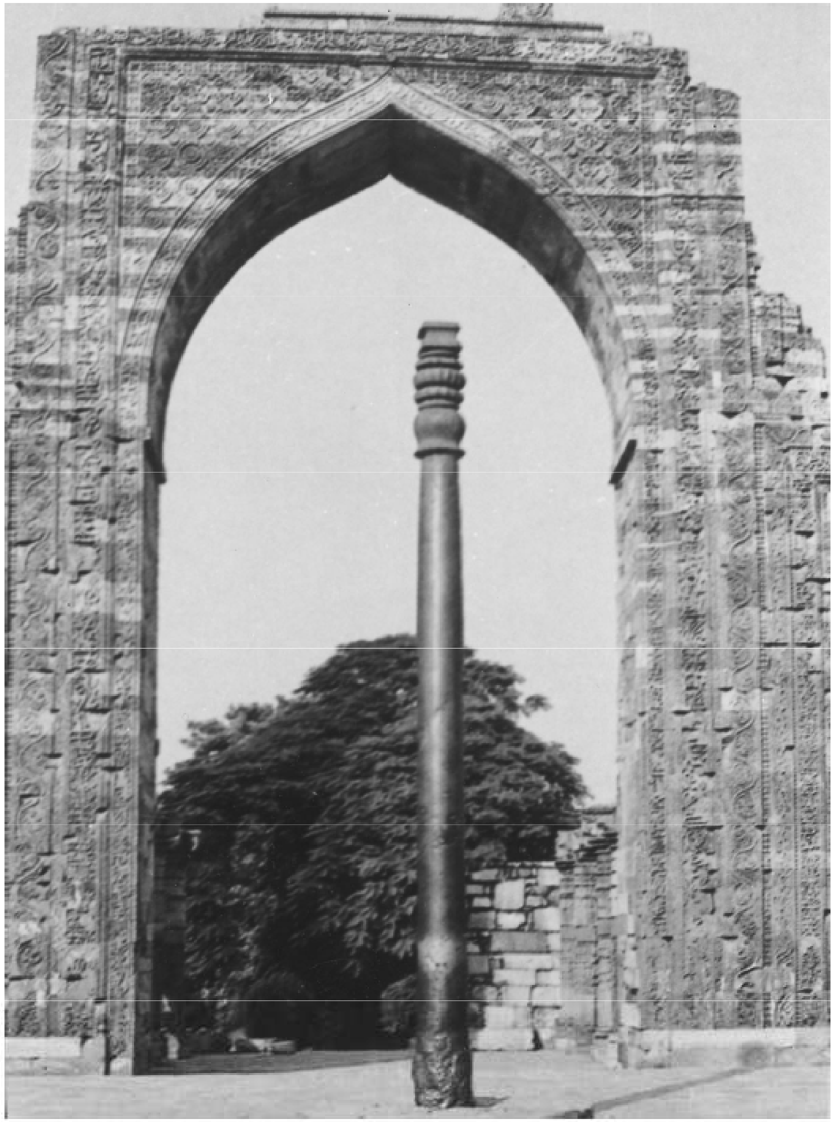


Fig. 1. The Delhi iron pillar before the construction of the iron grill cage around the stone platform.

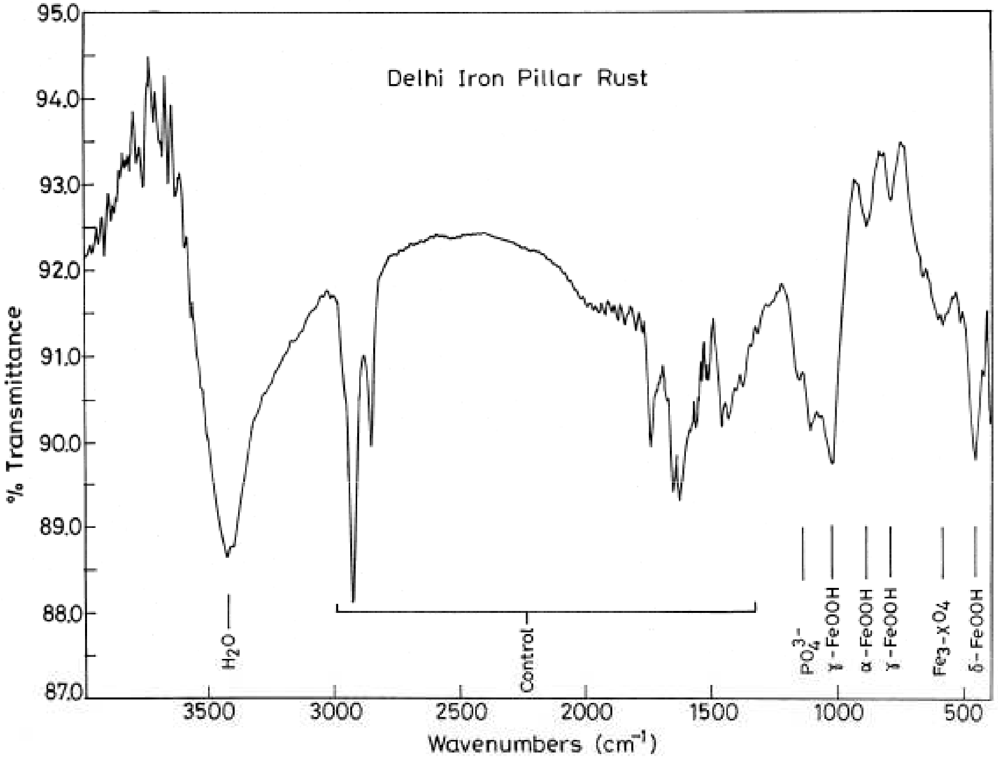


Fig. 5. FTIR spectrum from the DIP rust.

FTIR mikroskopie

Fig. 10.11. Egyptian bronze solid cast statuette of the God Osiris, inlaid with gold and blue glass. The surface has been extensively altered to massive light blue and dark green corrosion, identified as an overall patina of atacamite, with patches of chalconatronite. Frontal view. (*For a colored version of this figure, see Plate 10.III.*)

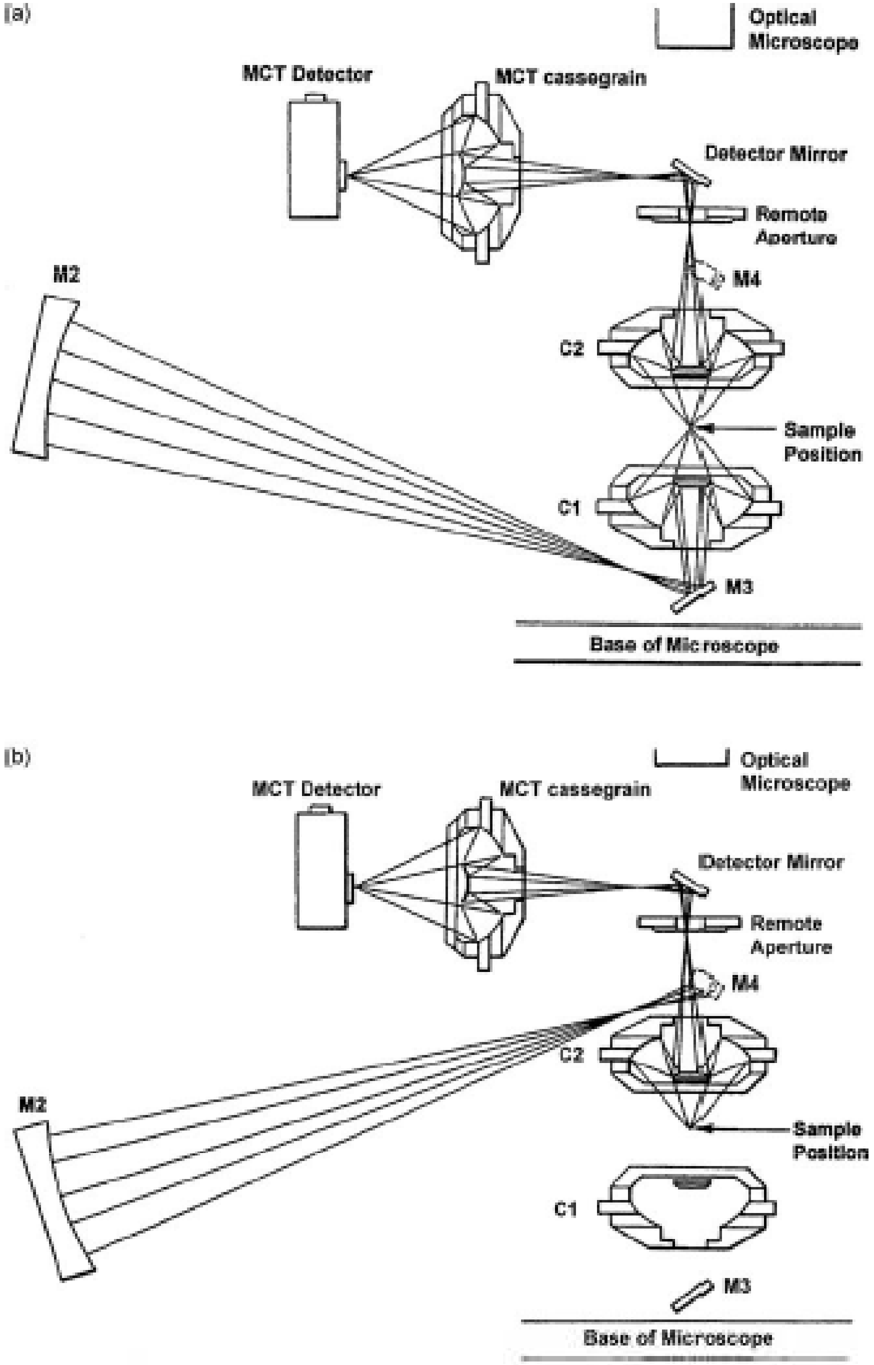


Figure 9.25 Optical paths of FTIR microscope with IR radiation: (a) transmittance; and (b) reflectance. M, mirror; C, Cassegrain lens. (Reproduced by permission of PerkinElmer Inc.)

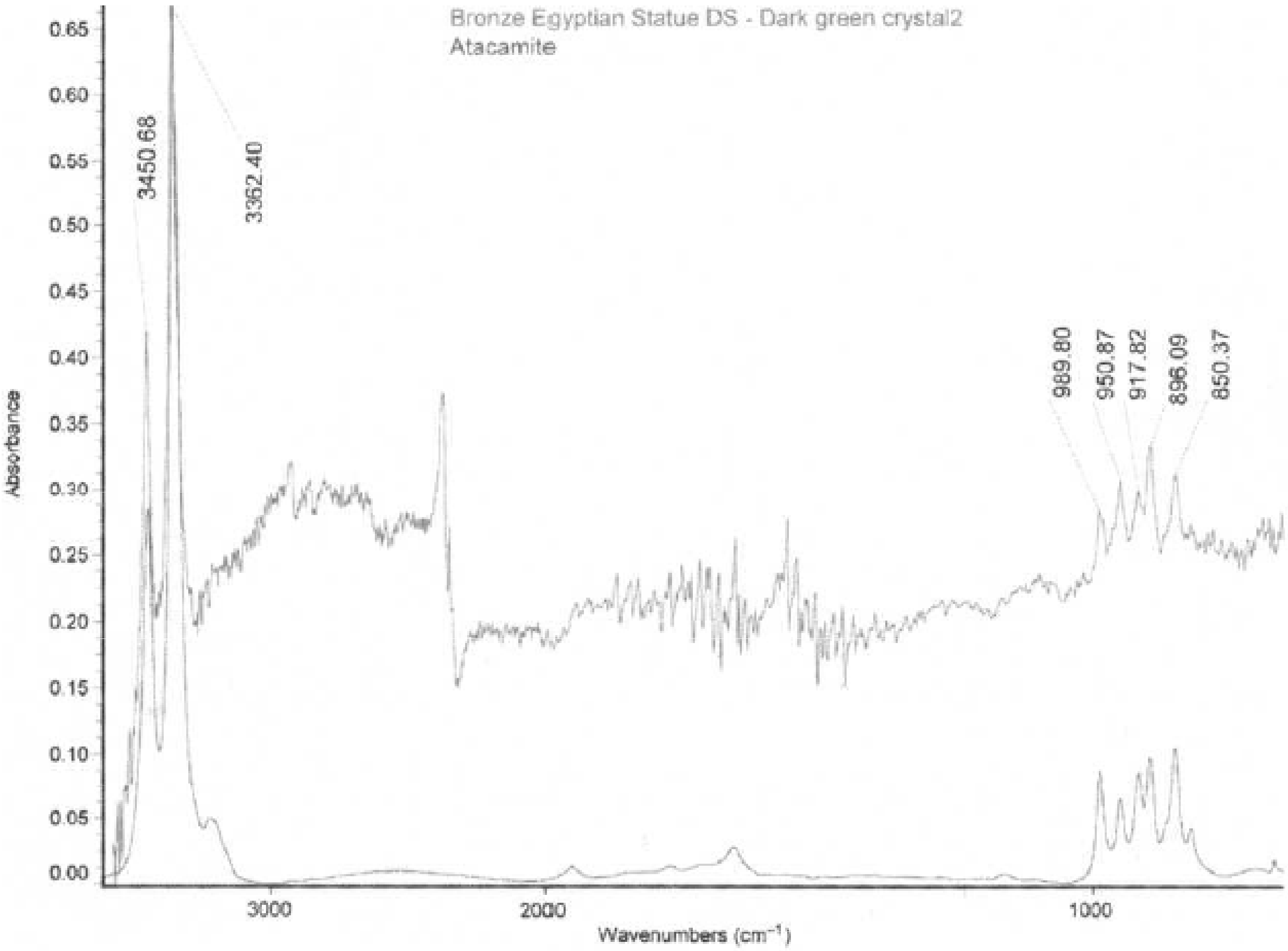


Fig. 10.17. FTIR Spectrum obtained, in situ, from the dark green surface of the Egyptian bronze Osiris shown in Fig. 10.11. The patina was shown to be of atacamite, but the result was only obtained with some difficulty and the chalconatronite patina could not be identified by this technique without sampling.

Infračervená spektrometrie

Terénní a mobilní zařízení

Reflektografie

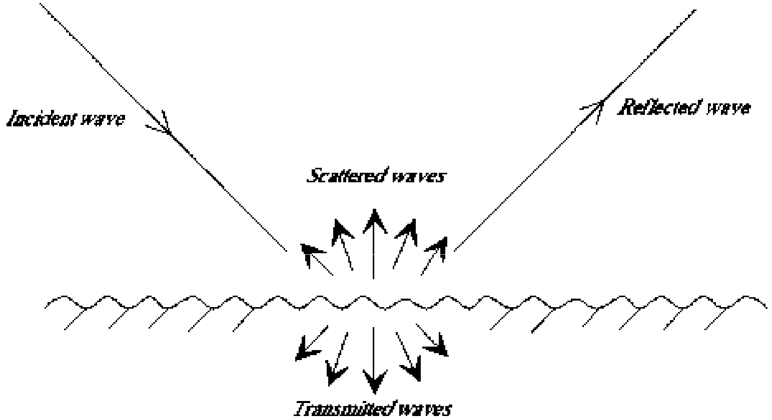


Figure 15. Case of natural environments: sum of specular reflection and diffuse reflection (volume reflection is ignored here).

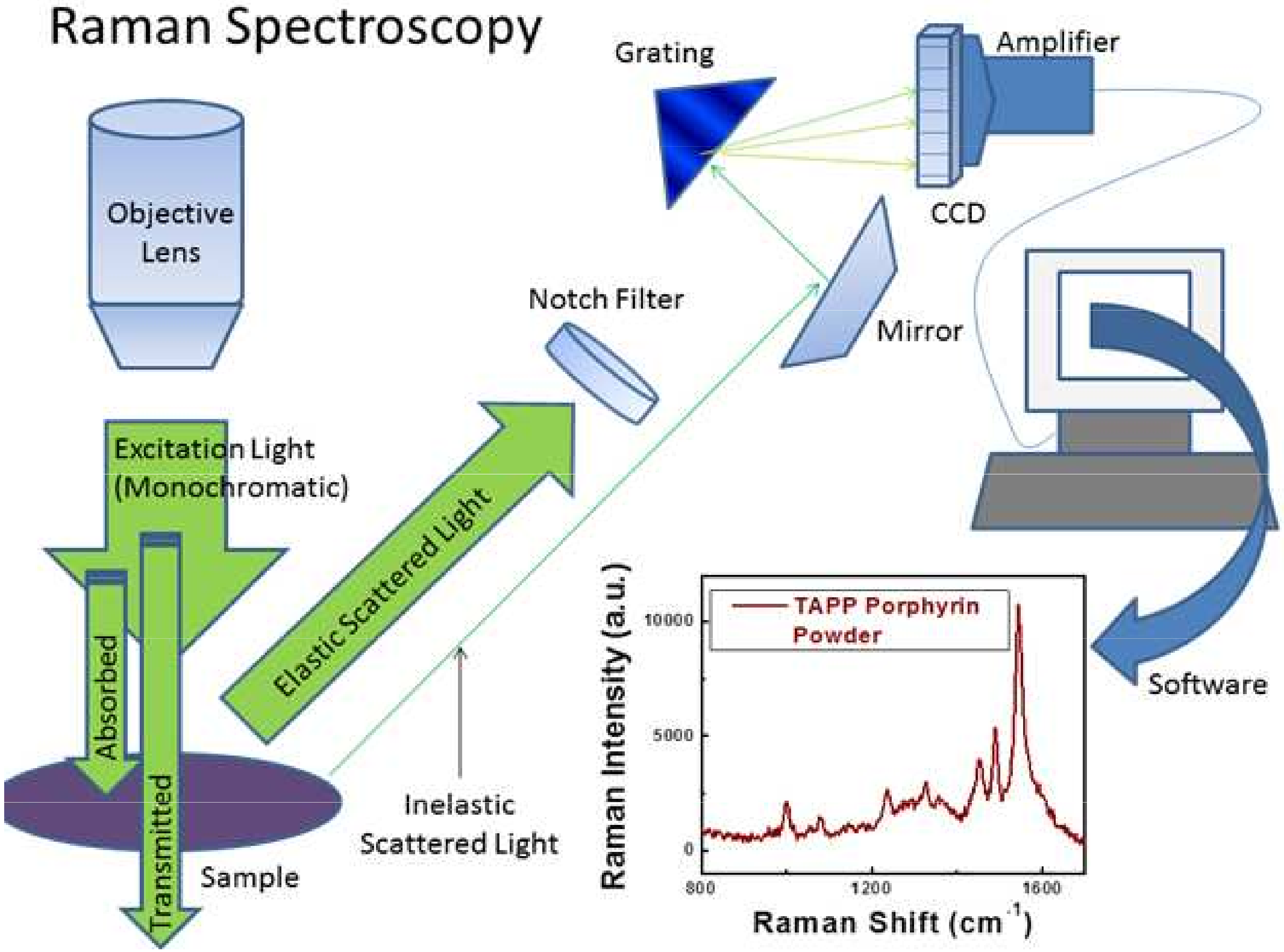
Aplikace infračervené reflektografie - zviditelnění podkresby

Reflektografie

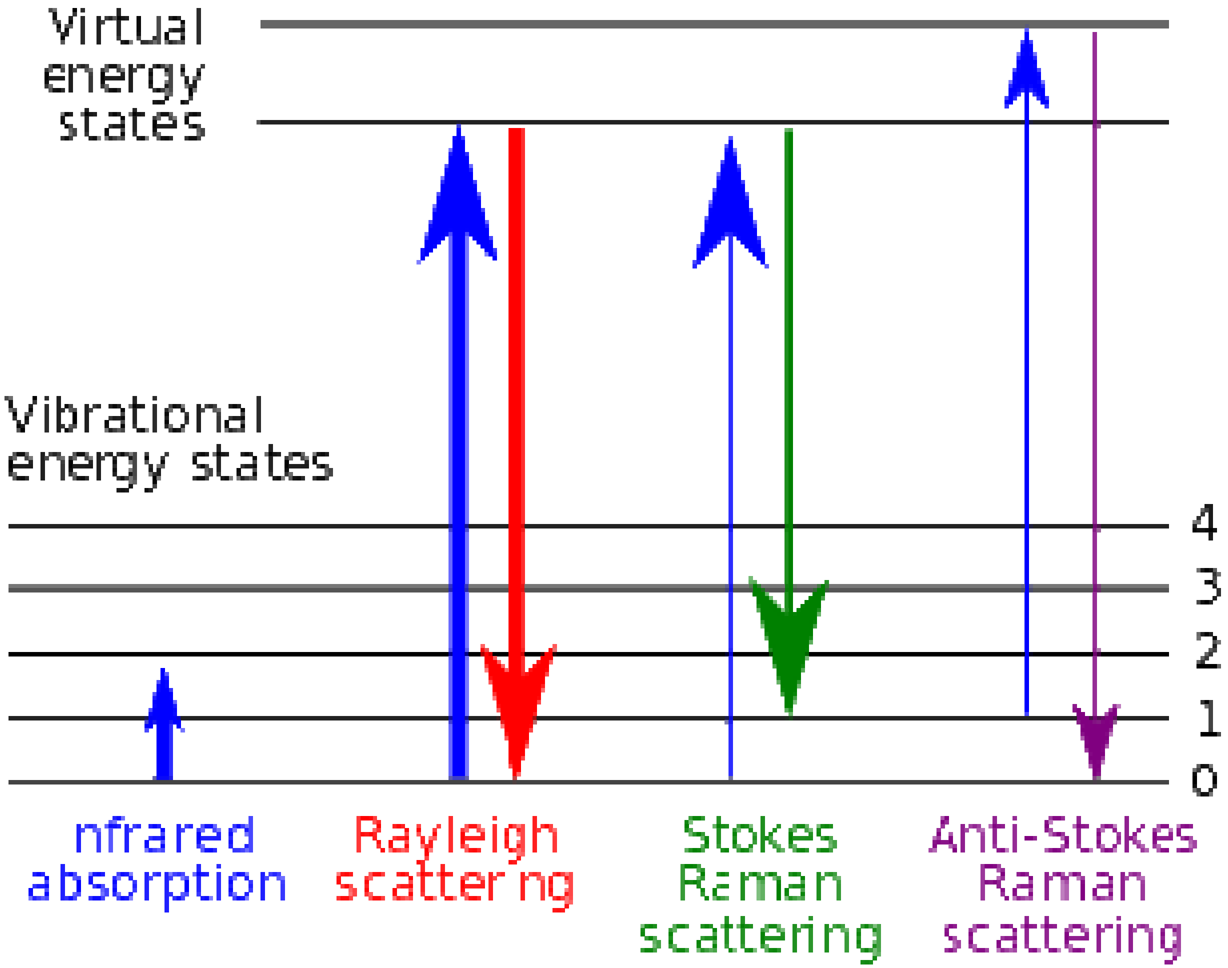
Aplikace infračervené reflektografie - zviditelnění tetování na mumifikovaných rukou z pohřebiště Semna South, Núbie (dnešní Súdán), stáří cca 2000 let.

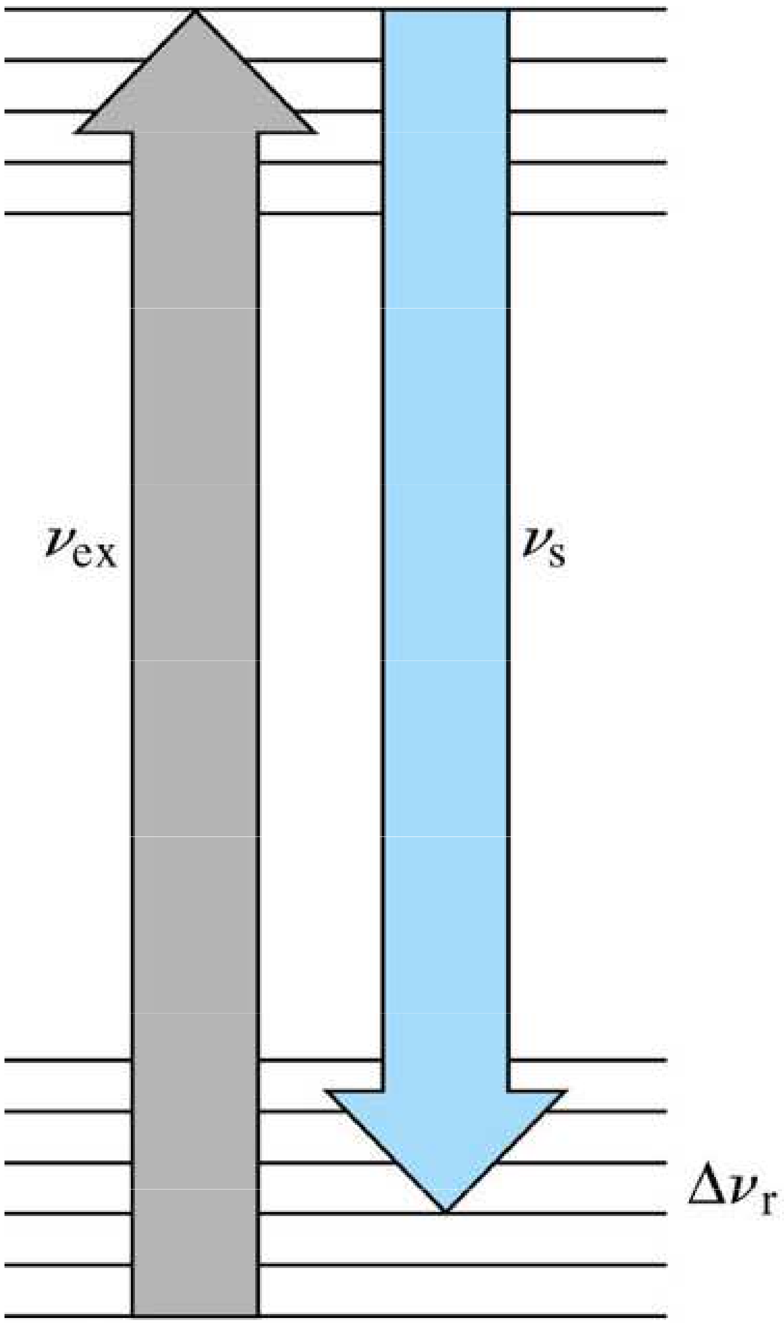
Ramanova spektrometrie

Nd:YAG 1064 nm
He–Ne 632.8 nm



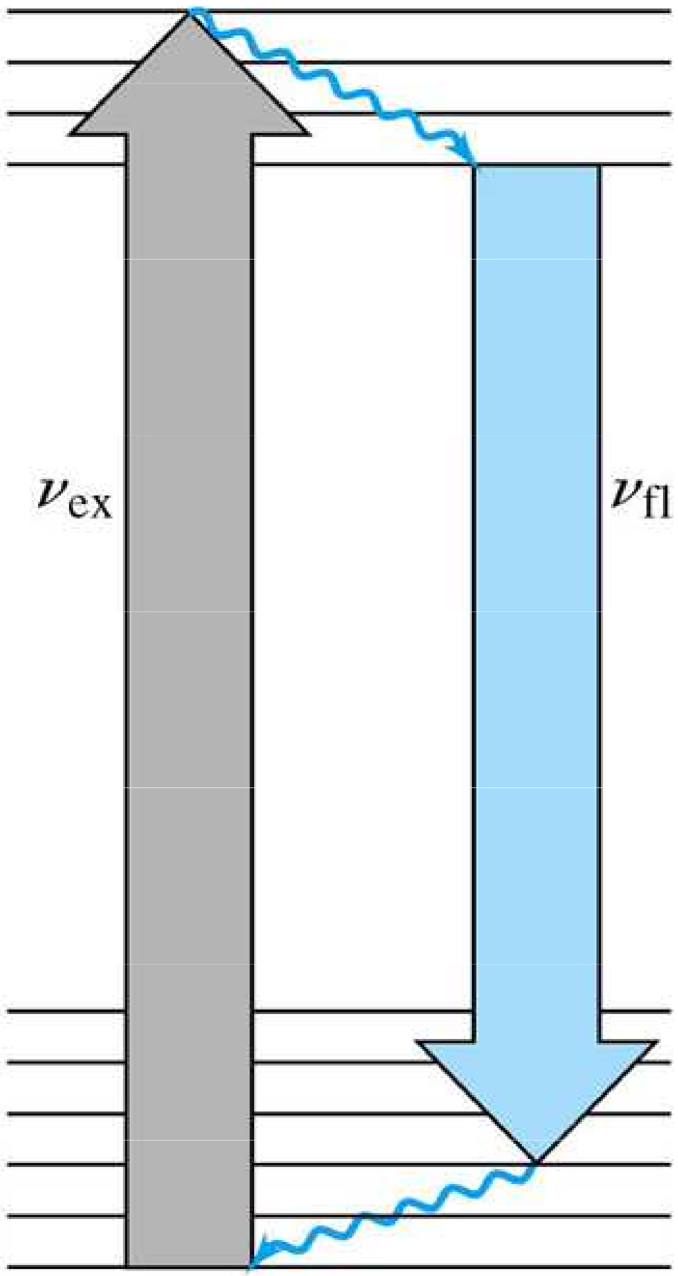
Princip Ramanovy spektrometrie





Resonance Raman

(a)



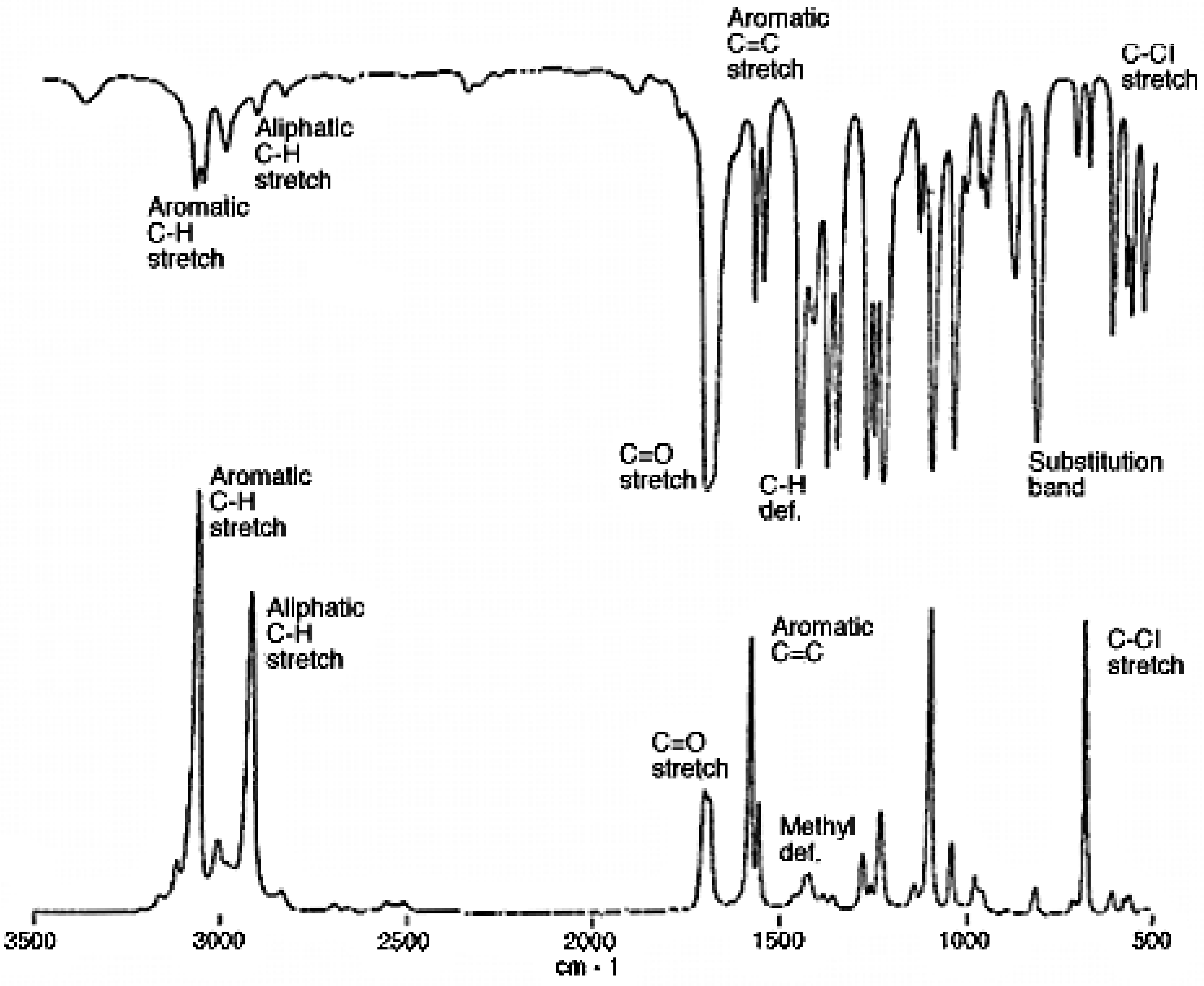
Fluorescence

(b)

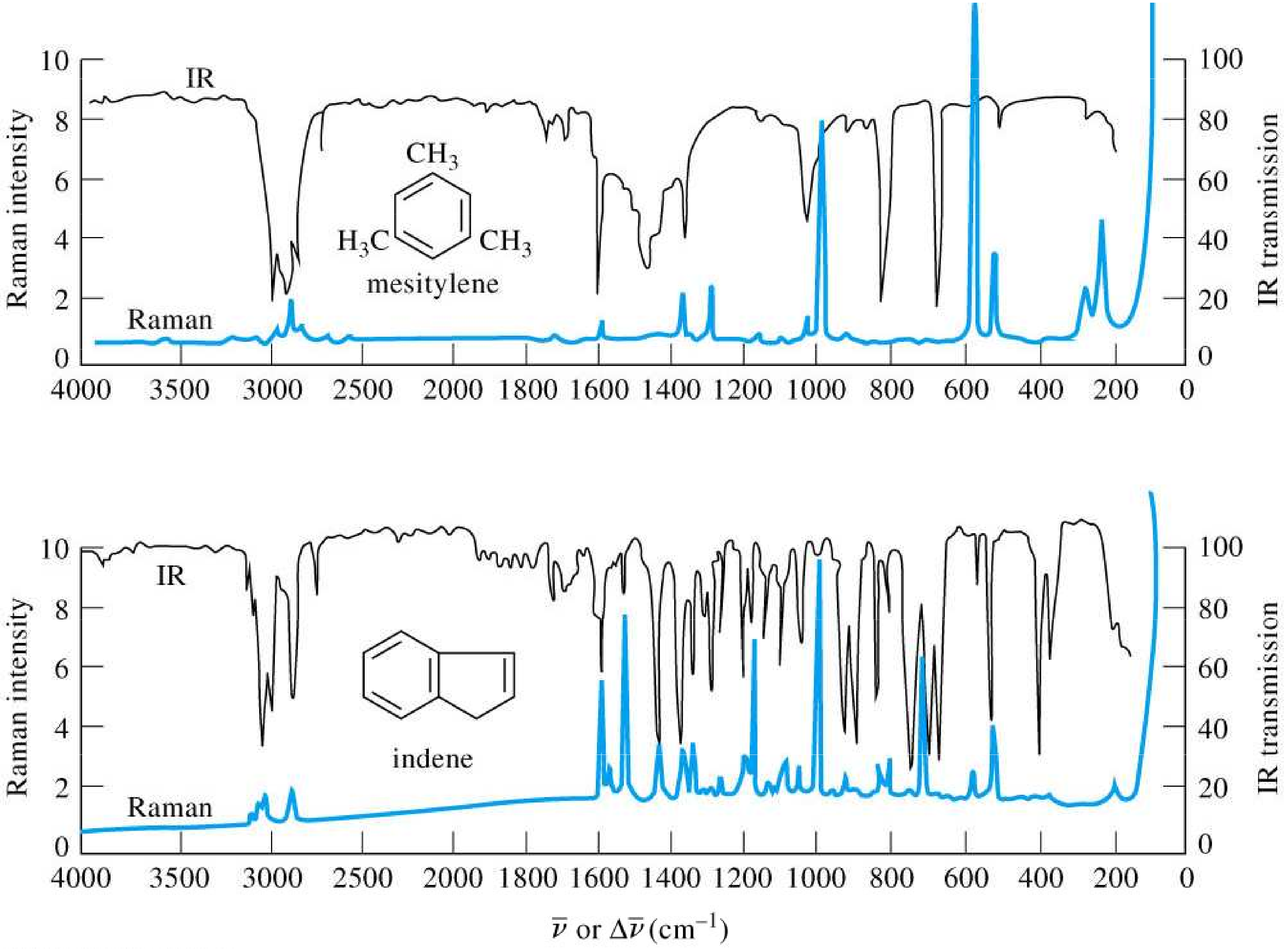
Raman vs. FTIR

FT IR

(FT) Raman



- comparison of Raman and IR Spectra



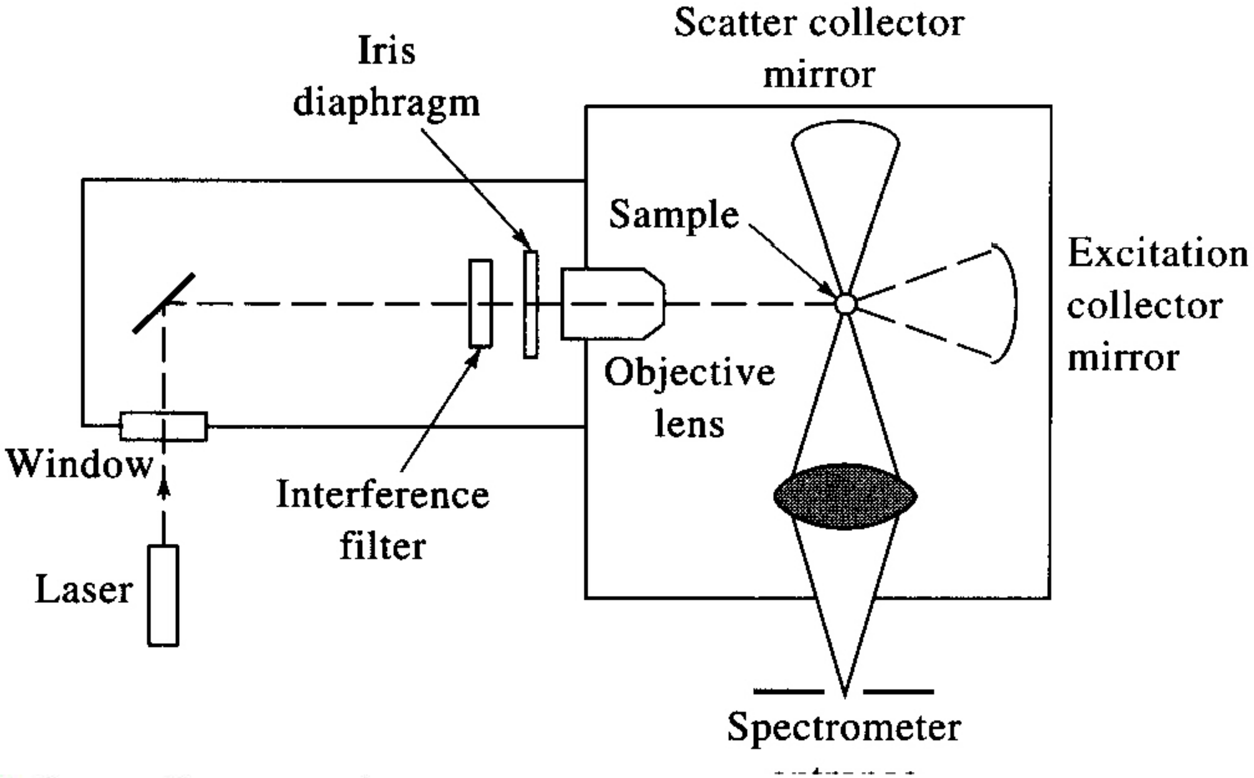
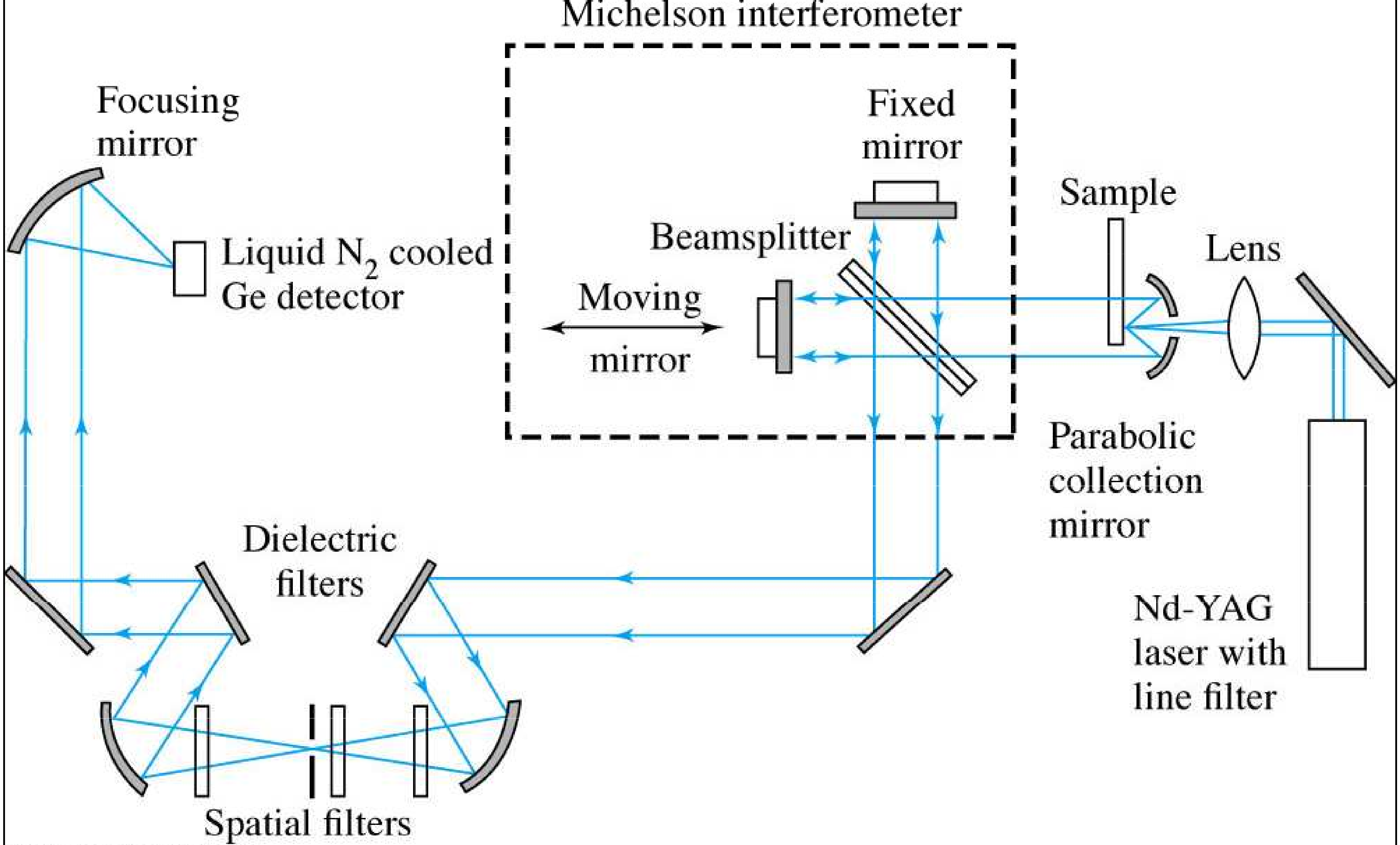


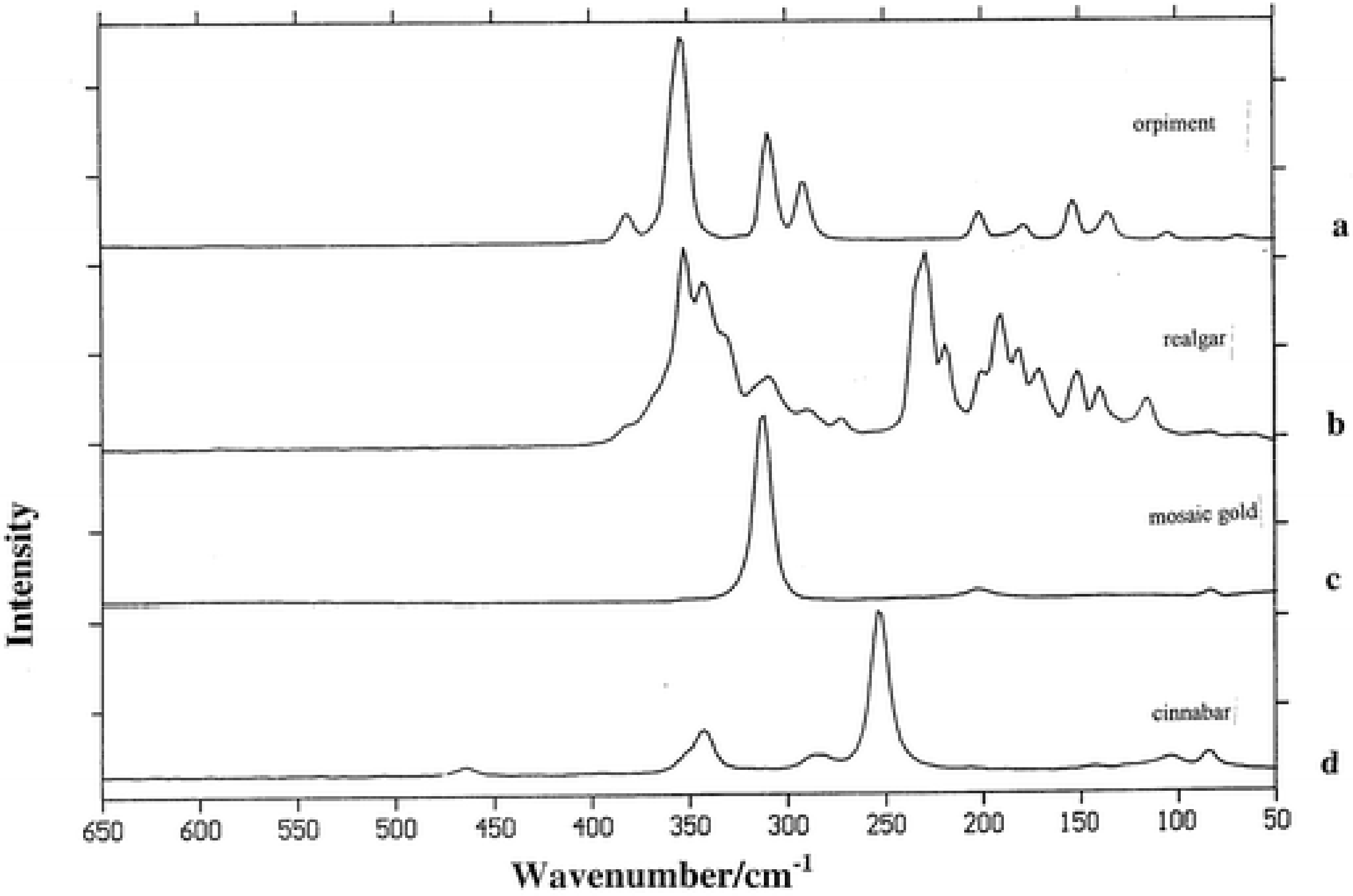
TABLE 18-1 Some Common Laser Sources for Raman Spectroscopy

Laser Type	Wavelength, nm
Argon ion	488.0 or 514.5
Krypton ion	530.9 or 647.1
Helium-neon	632.8
Diode	785 or 830
Nd-YAG	1064


Michelson interferometer
Focusing mirror
Fixed mirror
Sample
Beamsplitter
Lens
Liquid N_2 cooled Ge detector
Moving mirror
Parabolic collection mirror
Dielectric filters
Nd-YAG laser with line filter
Spatial filters

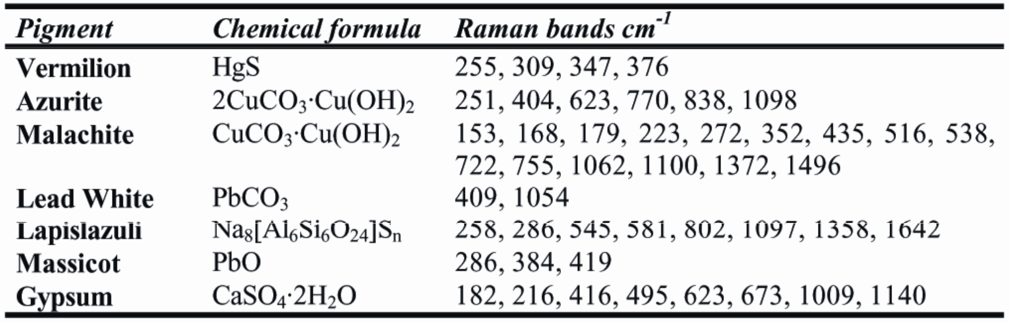
Ramanova spektrometrie pigmentů

Polychromovaná socha sv. Anny v Santa Maria la Real, Sasamon, Španělsko (13. stol.).



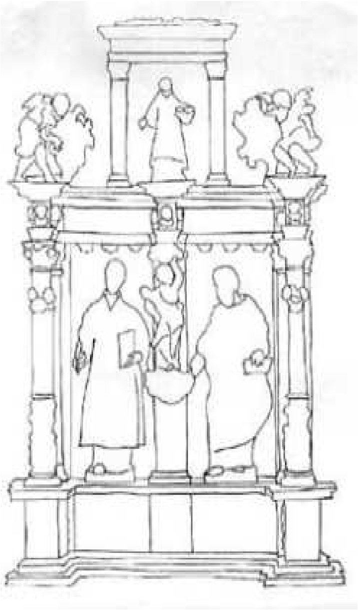
Ramanova spektra auripigmentu (As_2S_3), realgaru (As_4S_4), mozaikového zlata (SnS_2) a rumělky (Hg S).

Oltář ze "San Antolín y San Bernabé"



Pigment	*Chemical formula*	*Raman bands cm^{-1}*
Vermilion	HgS	255, 309, 347, 376
Azurite	$2CuCO_3 \cdot Cu(OH)_2$	251, 404, 623, 770, 838, 1098
Malachite	$CuCO_3 \cdot Cu(OH)_2$	153, 168, 179, 223, 272, 352, 435, 516, 538, 722, 755, 1062, 1100, 1372, 1496
Lead White	$PbCO_3$	409, 1054
Lapislazuli	$Na_8[Al_6Si_6O_{24}]S_n$	258, 286, 545, 581, 802, 1097, 1358, 1642
Massicot	PbO	286, 384, 419
Gypsum	$CaSO_4 \cdot 2H_2O$	182, 216, 416, 495, 623, 673, 1009, 1140

Table 1. Chemical formula and Raman band observed (in cm^{-1}, wavenumber) of the identified pigments.



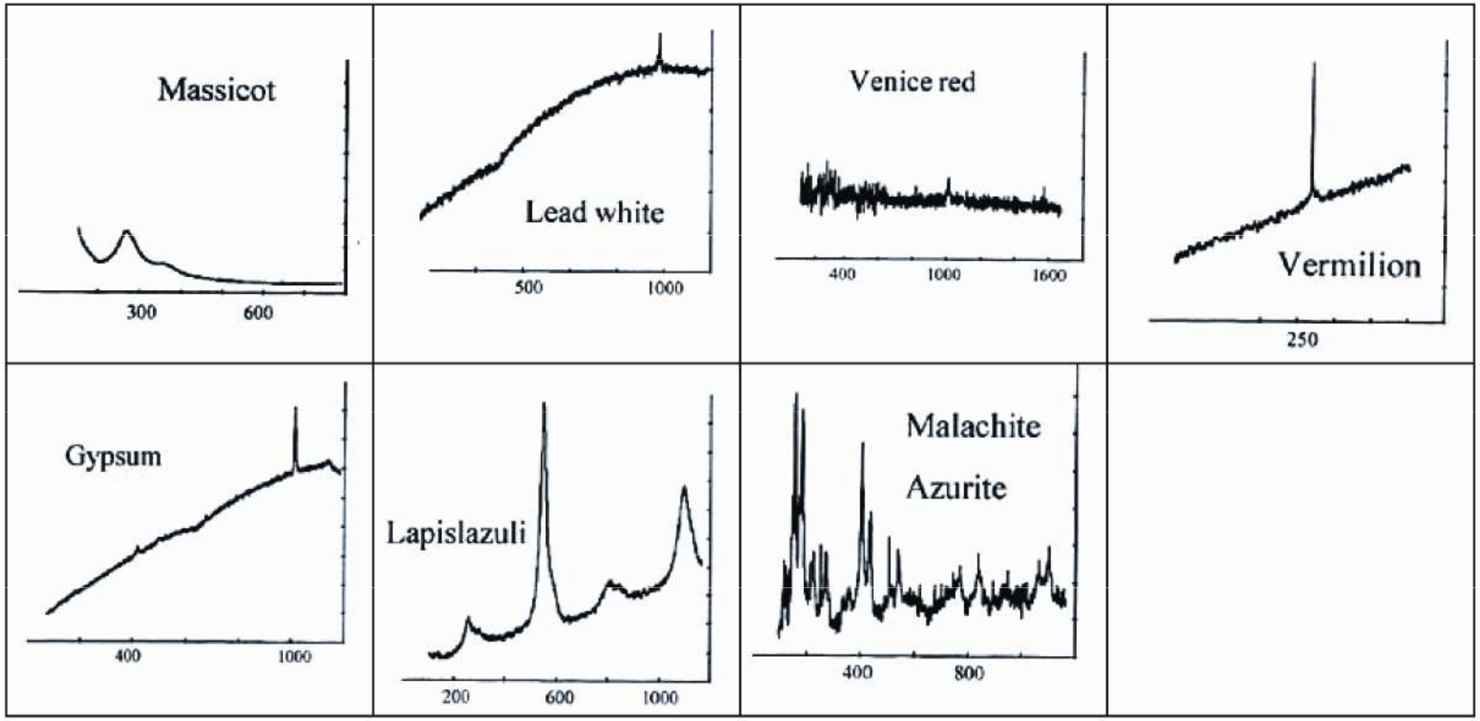


Fig. 5. The Raman spectra of the pictorial materials obtained from the "San Antolín y San Bernabé" altarpiece. The axis are Raman intensity and wavenumber (cm^{-1}).

Rozpoznání imitací

Přírodní barevné korály (karoten)

Barvené korály

Imitace korálů

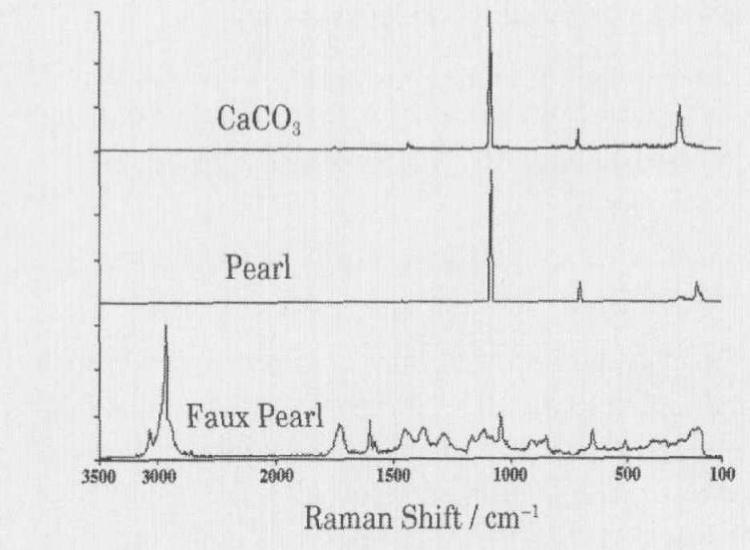
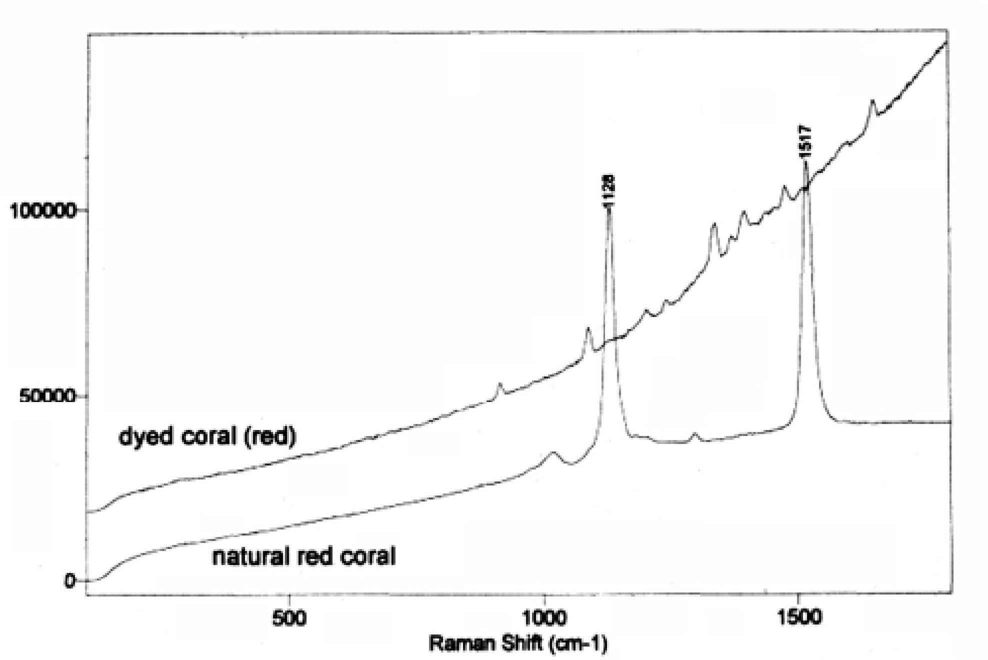


Figure 2. Raman spectra of CaCO$_3$, pearl, and faux pearl.



Identifikace slonoviny

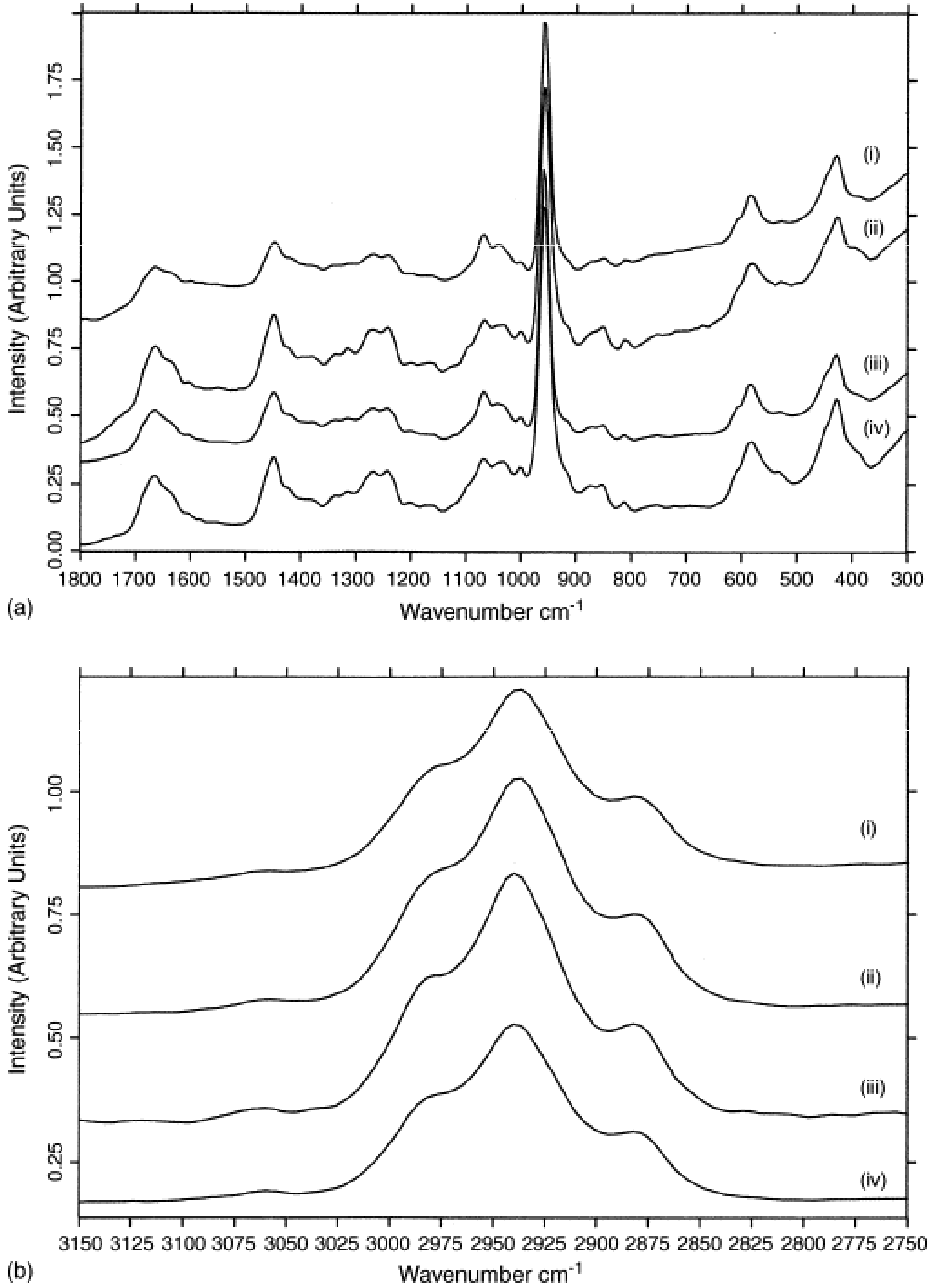
FT-Ramanova spektra:

Římské pečetidlo (i)

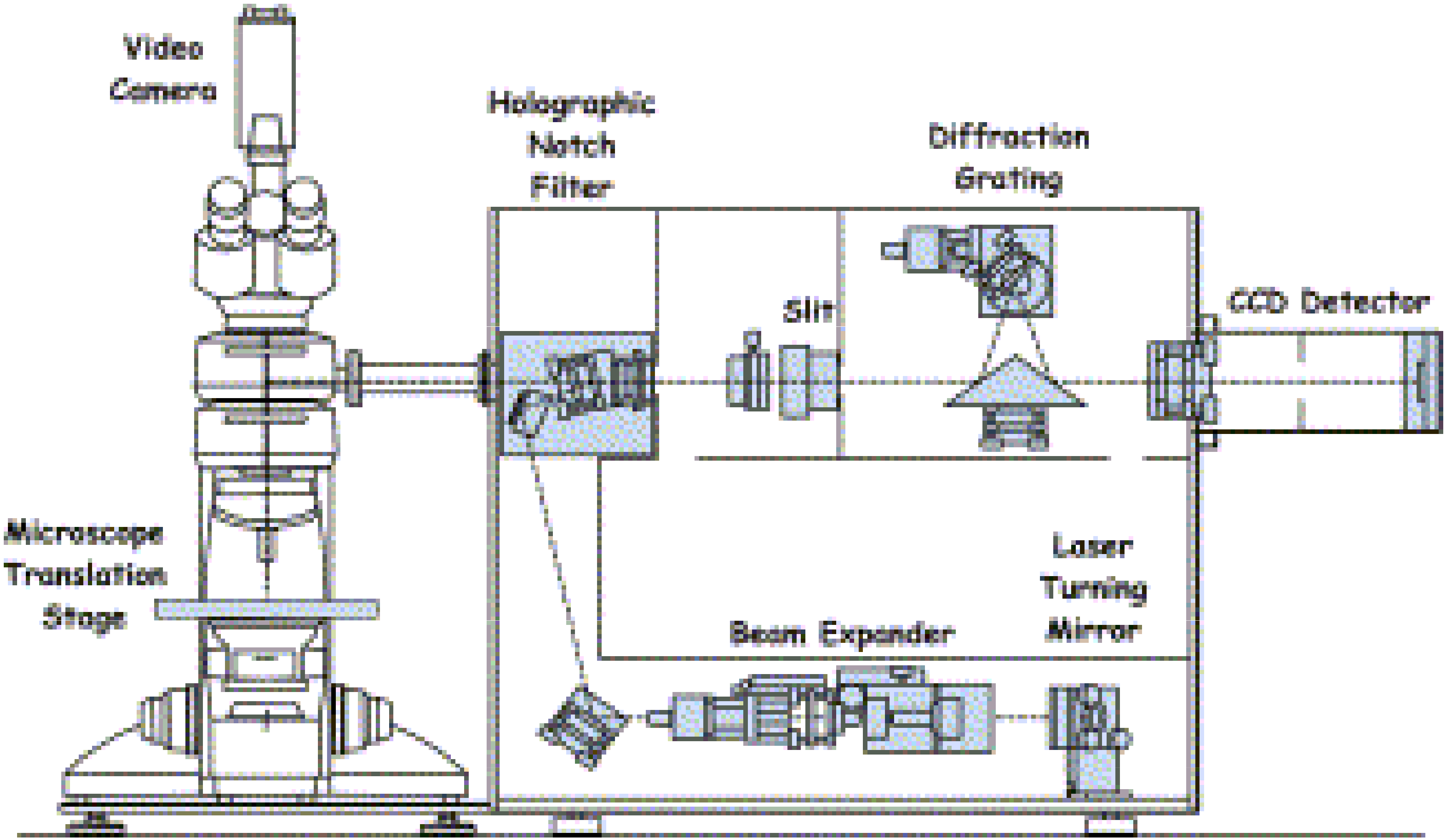
africký slon (ii),

vorvaň (iii)

hroch (iv)

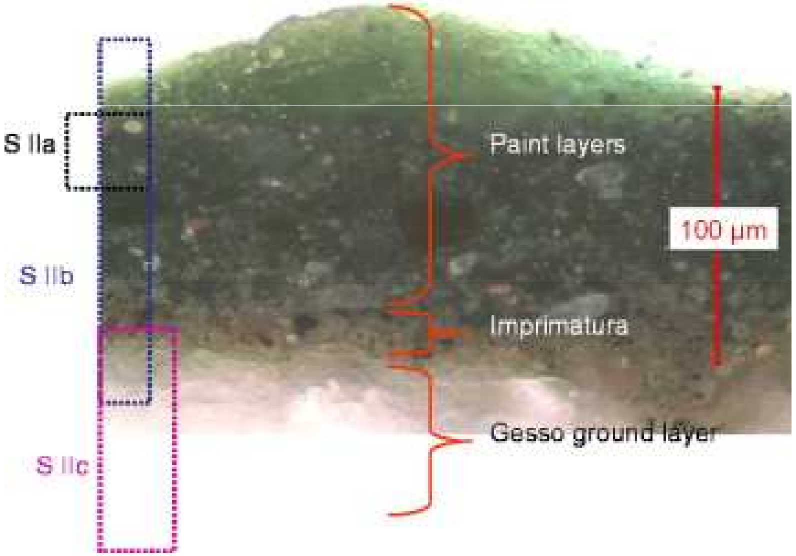


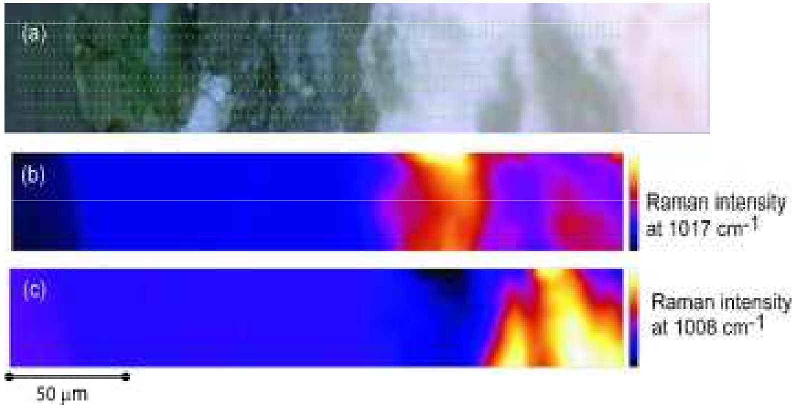
Ramanova mikroskopie



Ramanova mikroskopie maleb

***Portrét mladíka* (neznámý severoitalský malíř, cca 1515)**





Ramanovské mapy vzorku S IIb.
(a) optický obraz, (b) anhydrit, (c) sádrovec (gypsum).

Ramanova mikroskopie

mikrofosilie
v jurských
rohovcích

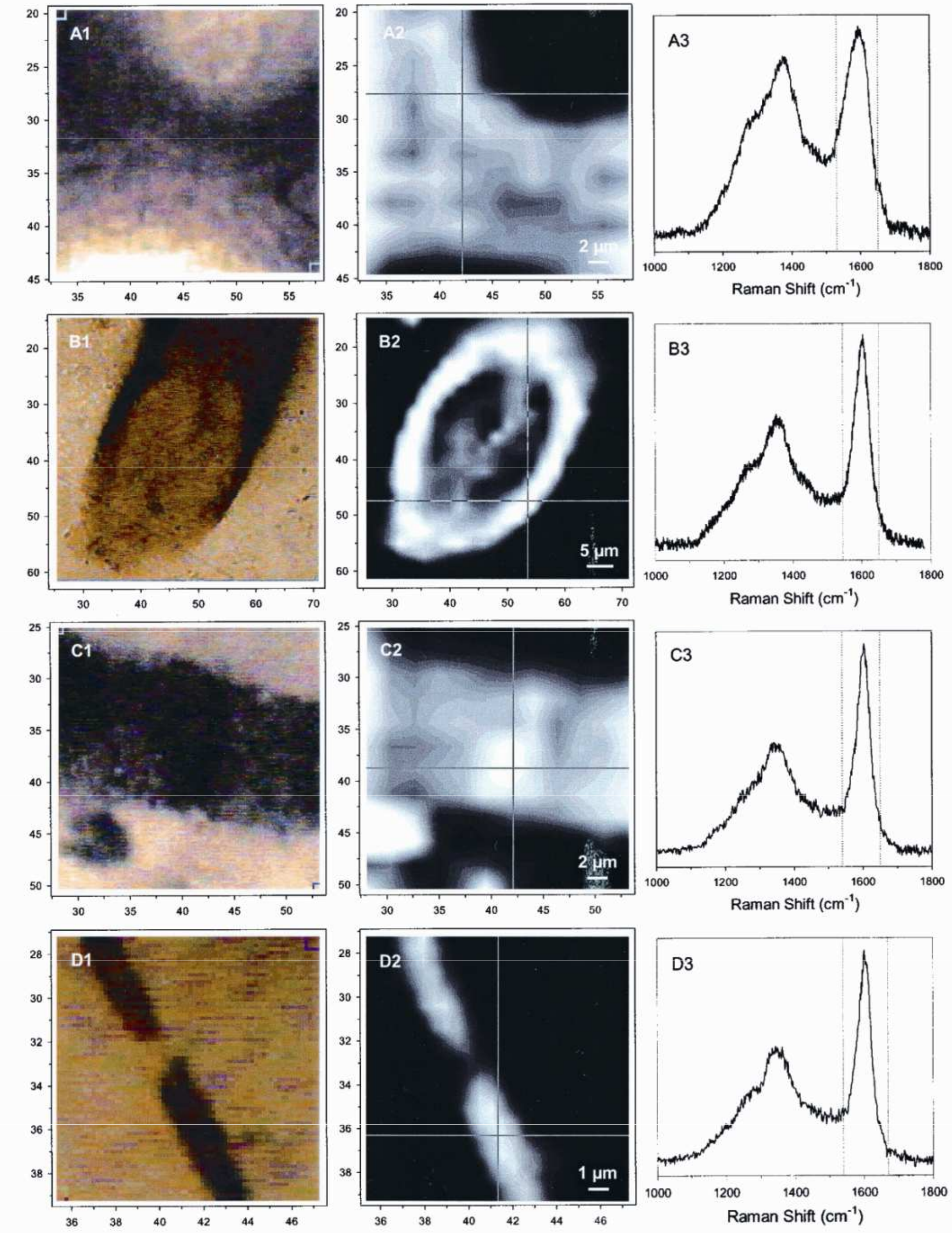
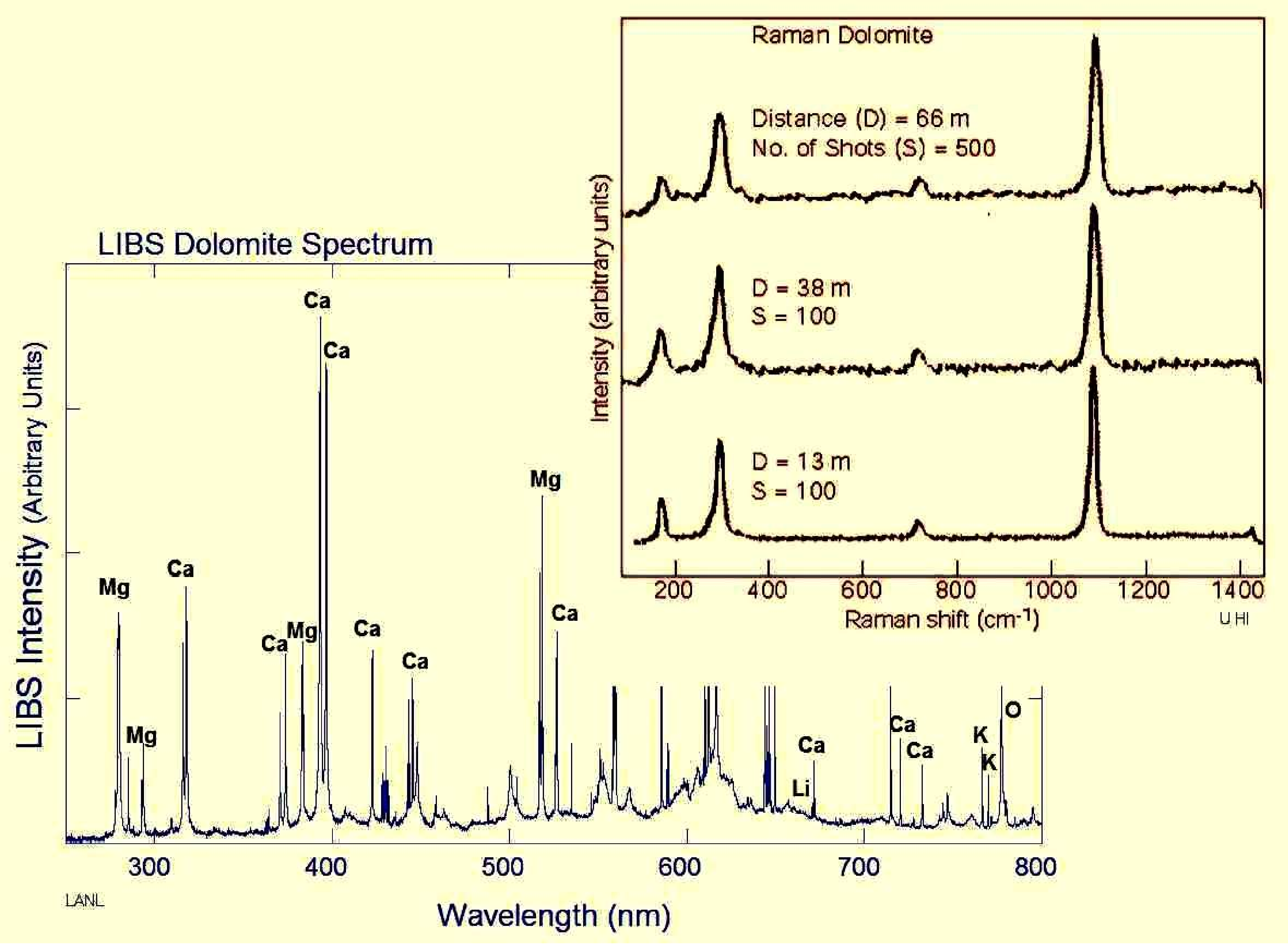


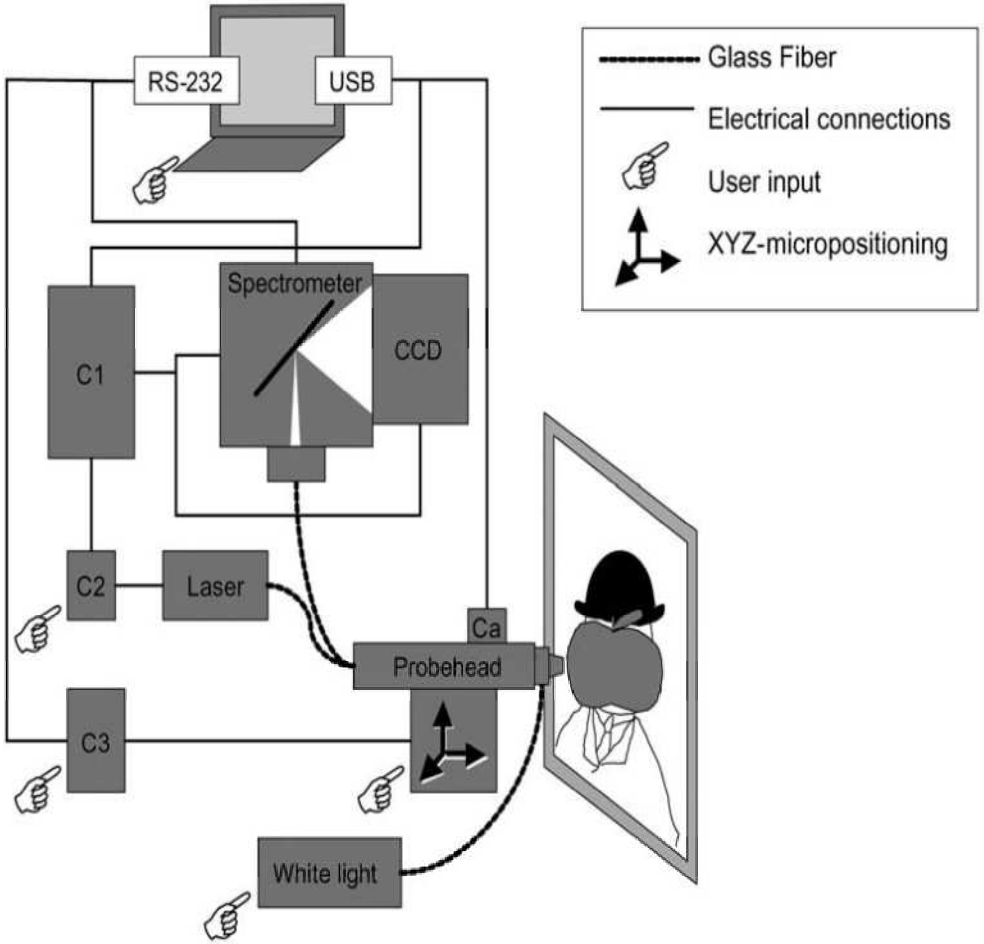
Fig. 1. Optical images (column 1), Raman images (column 2), and spectral bands used for Raman imaging (column 3) of permineralized carbonaceous fossils at or near the upper surfaces of polished chert thin sections: (*A*) Cell wall in the conductive tissue (lignified xylem) of an aquatic fern cf. *Dennstaedtia* from the essentially unmetamorphosed ≈45-Ma-old Clarno Formation of Oregon. (*B*) Tangential section of the tubular sheath of a *Lyngbya*-like oscillatoriacean cyanobacterium in a conical stromatolite (*Conophyton gaubitza*) from the subgreenschist facies ≈650-Ma-old Chichkan Formation of Kazakstan. (*C*) Transverse cell wall of a broad cellular trichome (*Gunflintia grandis*), and (*D*) a narrow prokaryotic filament (*G. minuta*), in domical stromatolites of the greenschist facies ≈2,100-Ma-old Gunflint Formation of Ontario, Canada. Each Raman image was produced by combining several hundred pixel-assigned point spectra (''spexels''), like those shown for each specimen in column 3, acquired over a small square part of the total area analyzed. The resolution of the Raman images is defined by the pixel dimensions of their component spexels; for *A*–*C*, 2 µm per pixel, and for *D*, 0.5 µm per pixel.

Raman + FTIR mikroskopie

Kombinace Raman + LIBS



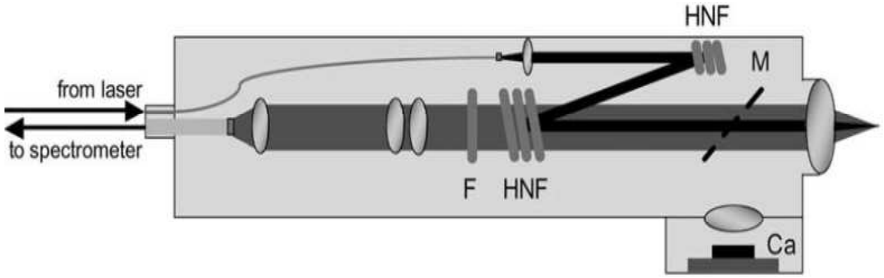
Mobilní zařízení pro Ramanovu spektrometrii



M : 95% propustné zrcadlo pro vizualizaci plochy kamerou (*Ca*).

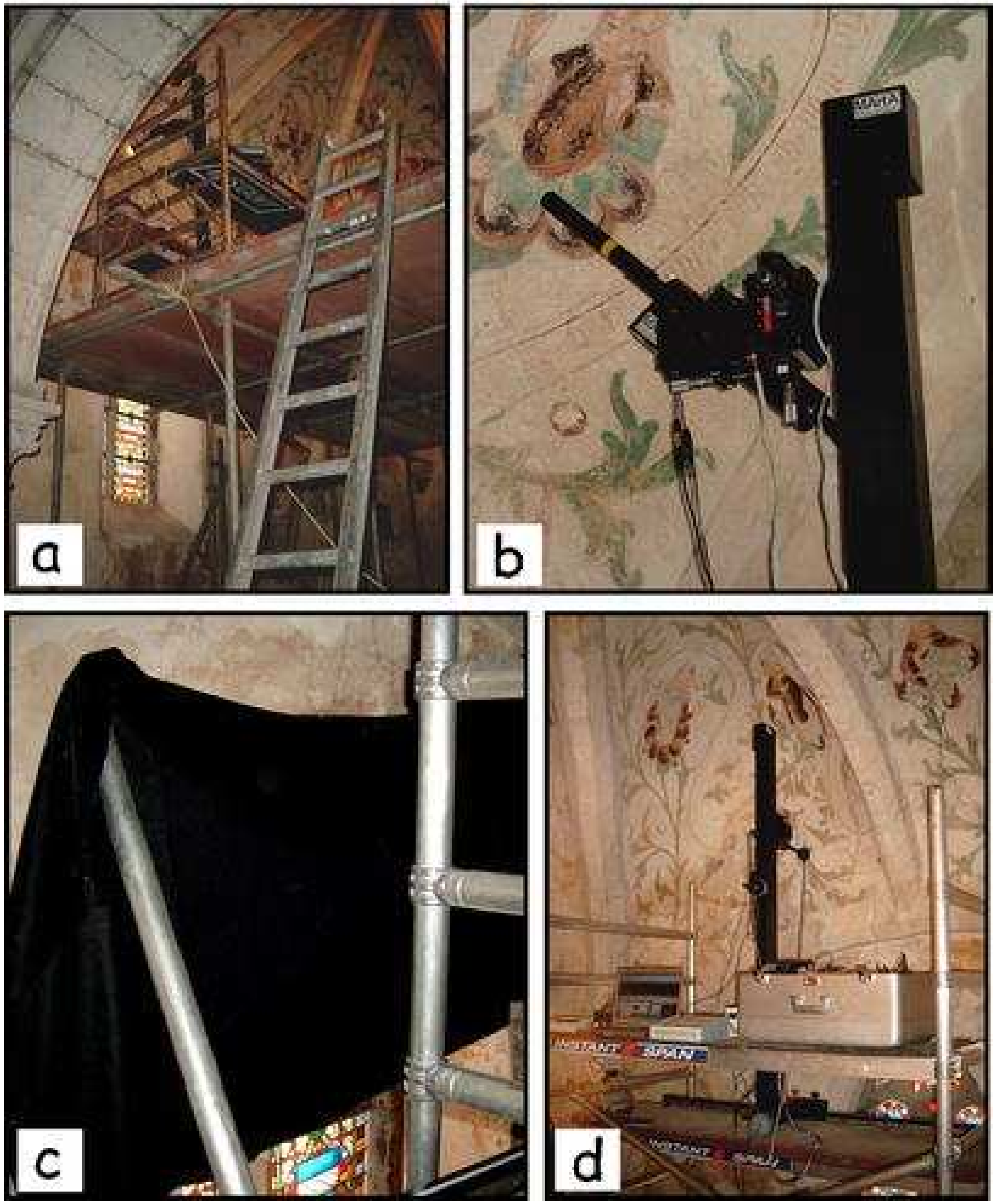
HNF : holografické filtry odrážející laserový paprsek a propouští ramanovsky posunuté záření (Stokes)

F filtry pro anti-Stokesovskou část spektra



Ramanova spektrometrie

Terénní a mobilní zařízení



Analýza nástěnných maleb (kaple Ponthoz.)

Ostatní metody

Rentgenová fluorescenční analýza (XRF)

Rentgenová

Radionuklidová

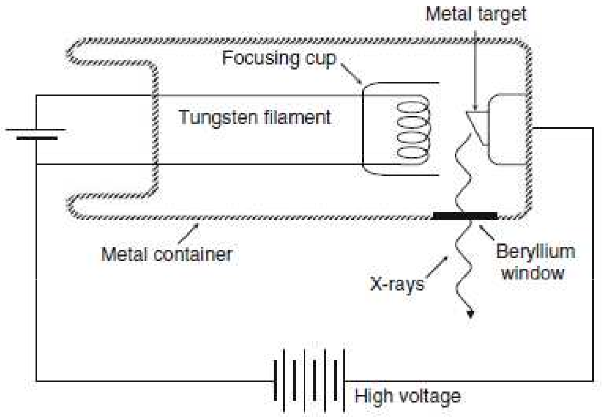


Fig. 37.1 X-ray tube ("Fundamentals of Physics", Second Edition Extended, David Halliday and Robert Resnick, copyright 1981, Reproduced with permission of John Wiley & Sons, Inc.)

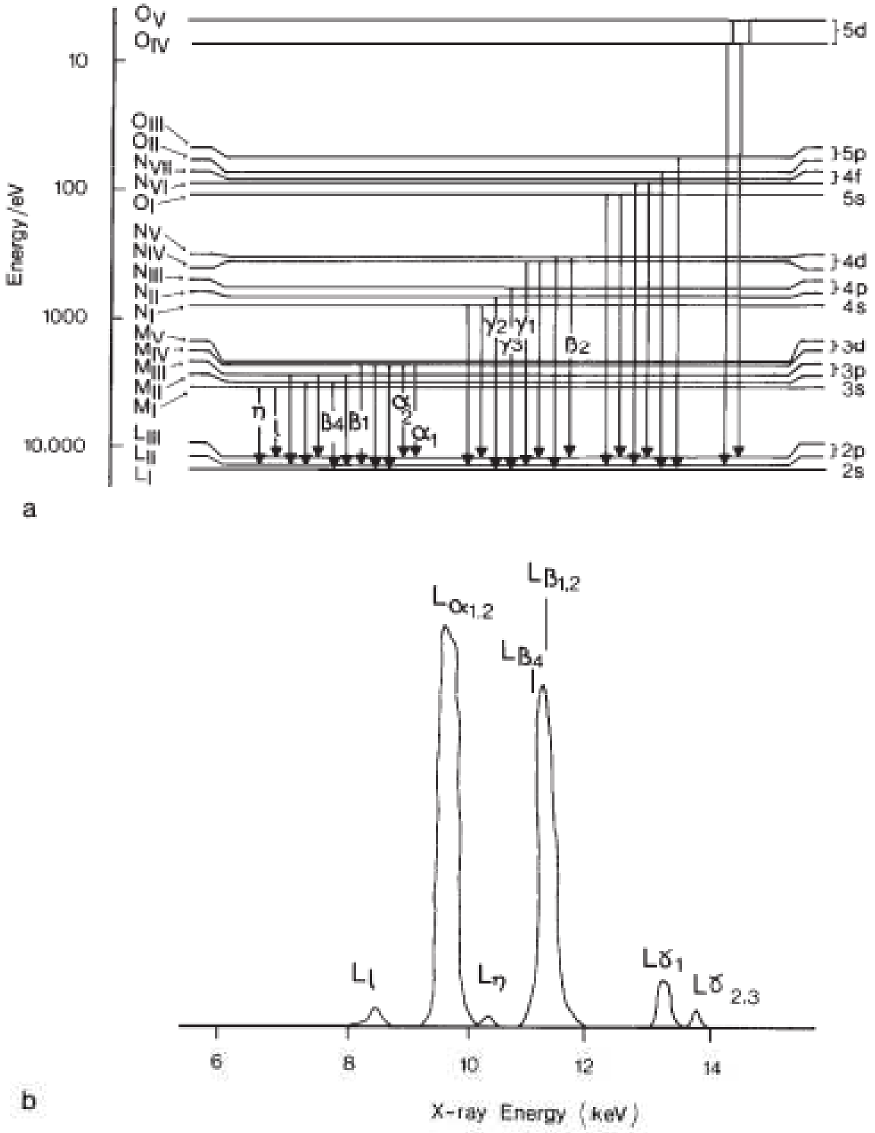


Figure 2.8 Electronic transitions giving rise to the L spectrum of gold. The L spectrum is considerably more complicated with three main lines normally resolved as shown in the accompanying spectrum – L_α (arising from two transitions), L_β (with up to 17 contributing transitions) and L_γ (up to 8 transitions), plus a number of 'forbidden' transitions. (After Jenkins, 1974; Fig. 2–11. © John Wiley & Sons Limited. Reproduced with permission.)

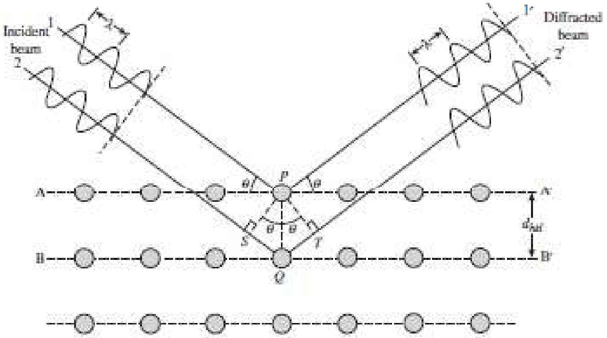


Figure 2.6 Bragg diffraction by crystal planes. The path difference between beams 1 and 2 is SQ + QT = 2 PQ sinθ. (Reproduced with permission from W.J. Callister Jr., *Materials Science and Engineering: An Introduction*, 7th ed., John Wiley & Sons Inc., New York. © 2006 John Wiley & Sons Inc.)

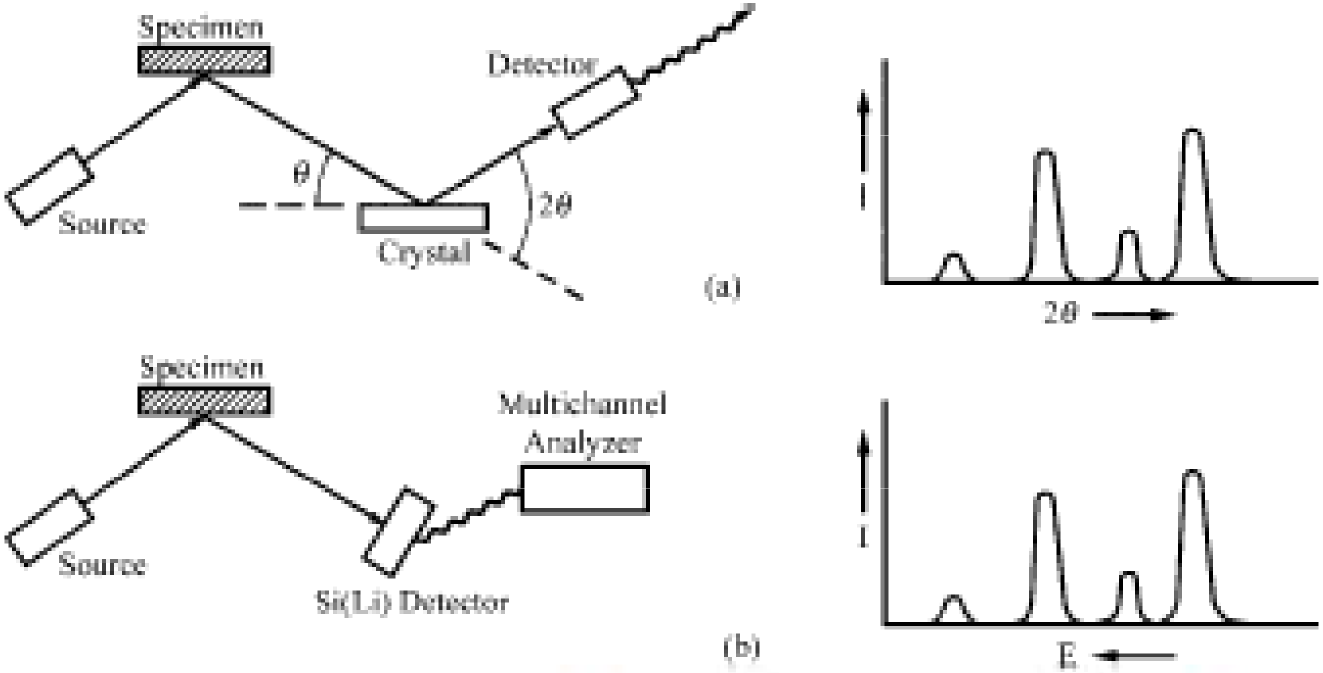
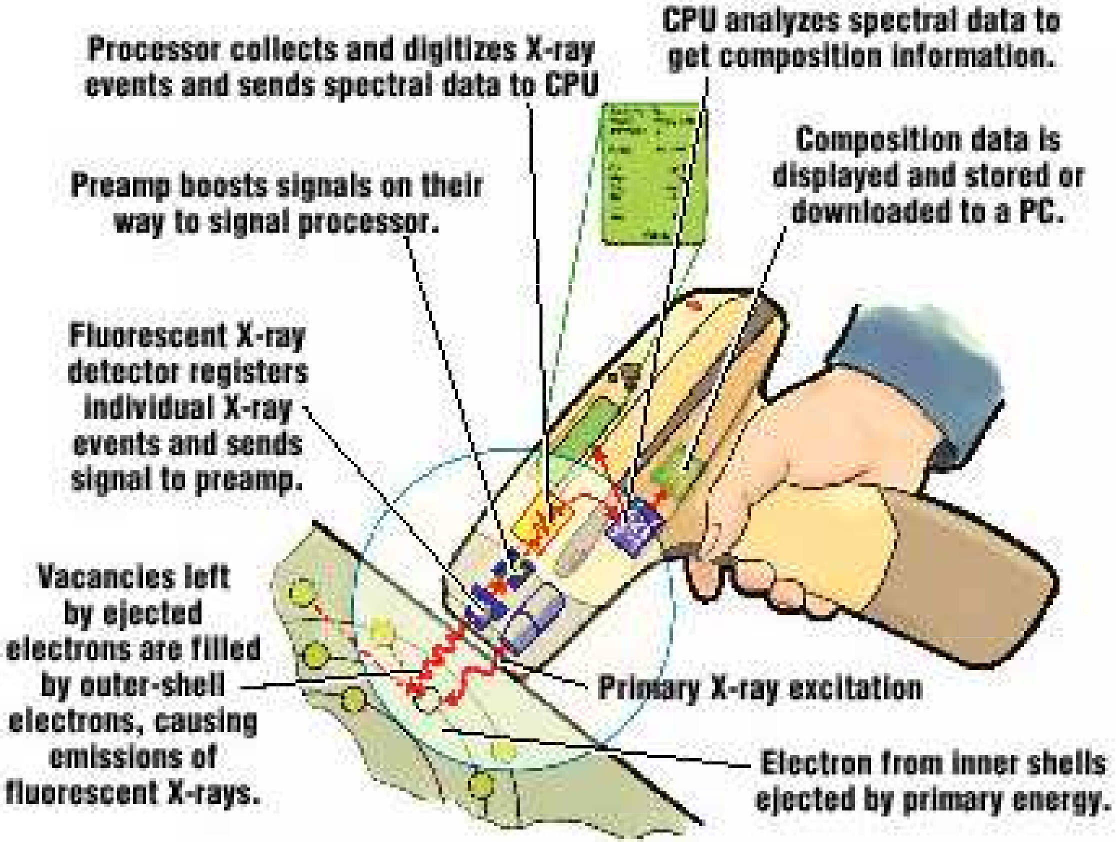


Figure 6.6 Main components and dispersive spectra of: (a) WDS; and (b) EDS.

Portable X-Ray Fluorescence (PXRF)



Processor collects and digitizes X-ray events and sends spectral data to CPU

CPU analyzes spectral data to get composition information.

Preamp boosts signals on their way to signal processor.

Composition data is displayed and stored or downloaded to a PC.

Fluorescent X-ray detector registers individual X-ray events and sends signal to preamp.

Vacancies left by ejected electrons are filled by outer-shell electrons, causing emissions of fluorescent X-rays.

Primary X-ray excitation

Electron from inner shells ejected by primary energy.

Elektronová mikrosonda

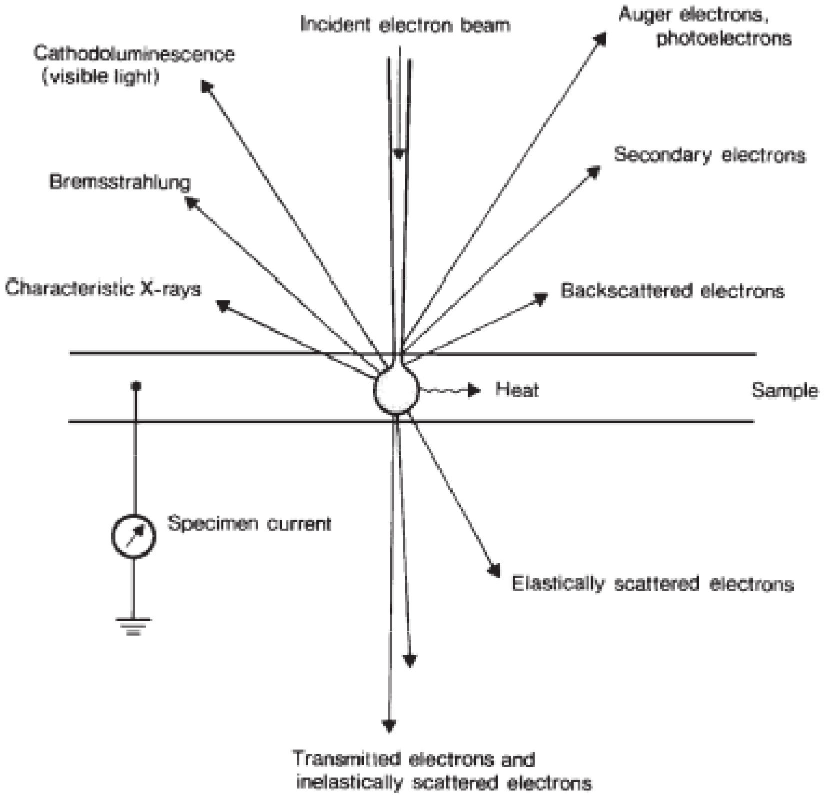


Figure 2.12 Interaction of primary electrons with a thin solid sample, showing the various processes which can take place. (After Woldseth, 1973; Fig. 4-1.)

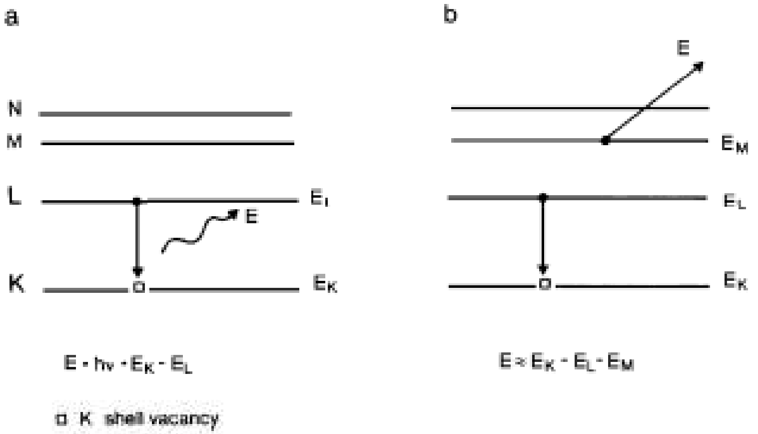


Figure 2.6 The X-ray emission and Auger processes. An inner vacancy in the K shell de-excites via one of two competing processes – (a) X-ray emission, in which an L electron drops down and the excess energy is carried away by an X-ray photon, or (b) the Auger process, in which an L electron drops down, but the excess energy is carried away by a third electron – in this case from the M shell.

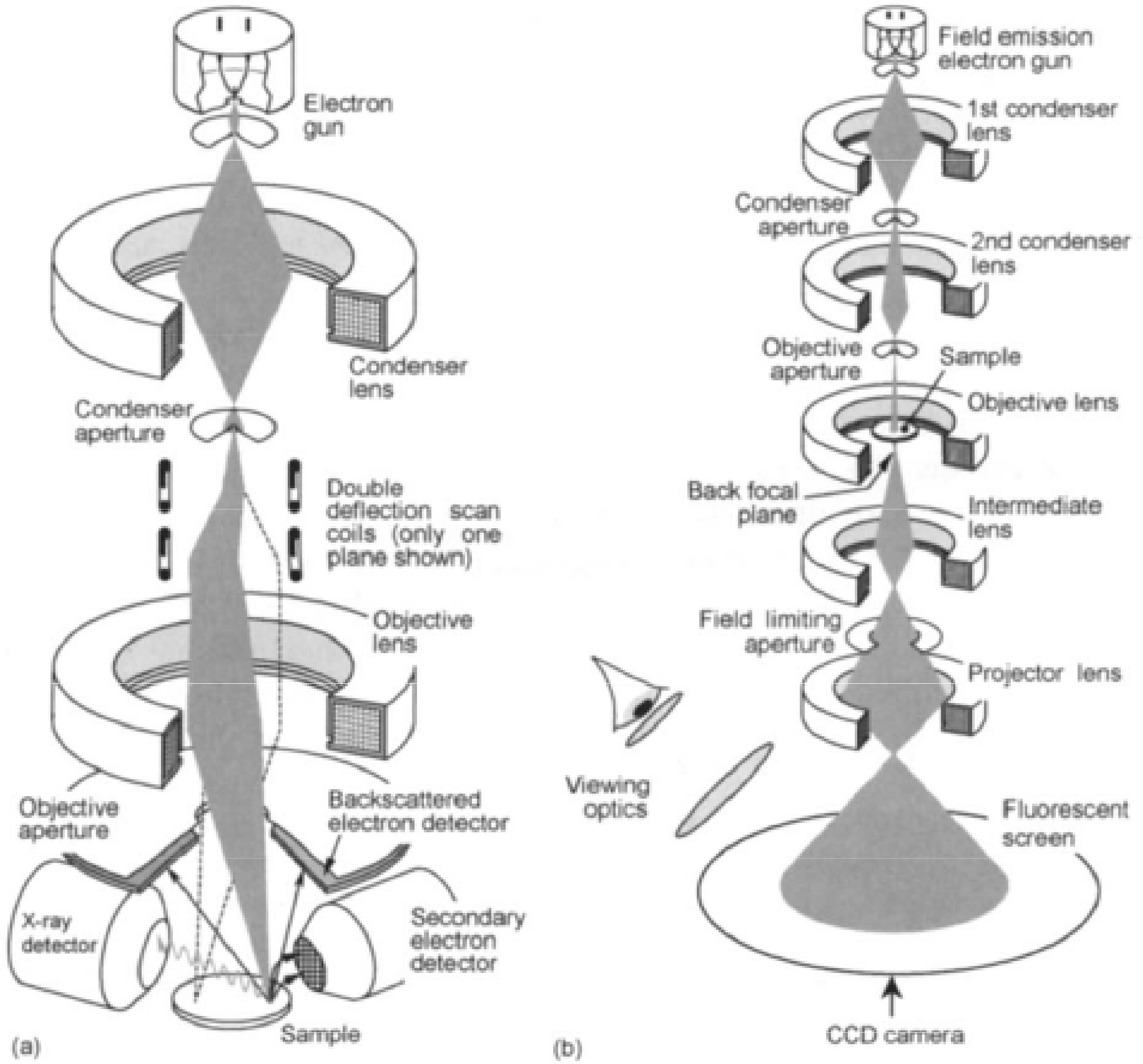


Fig. 3.1. General types of electron microscope. (a) Scanning electron microscope (SEM); (b) transmission electron microscope (TEM).

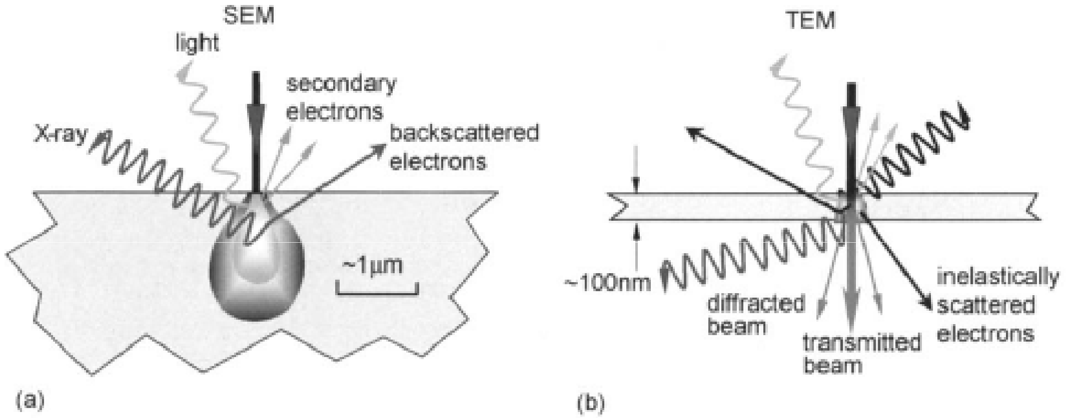


Fig. 3.2. Interaction of electrons with a solid showing effects of interaction volume. (a) SEM sample; (b) sample thinned for TEM.

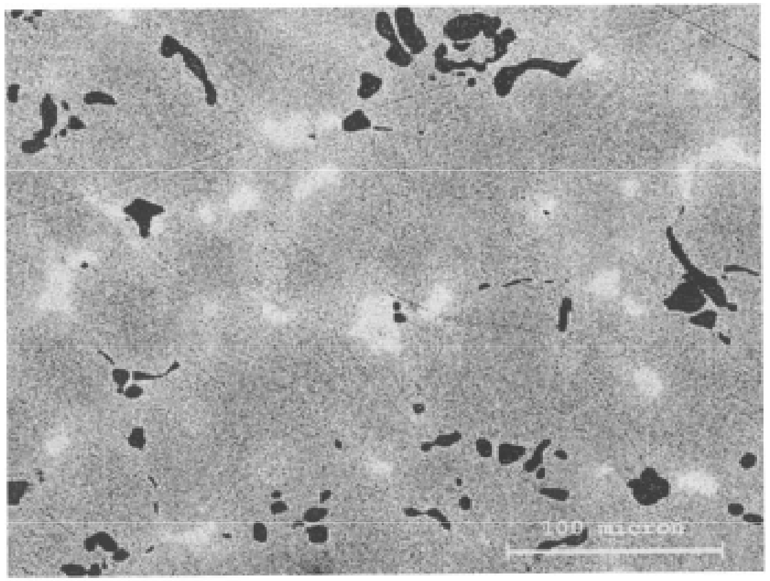
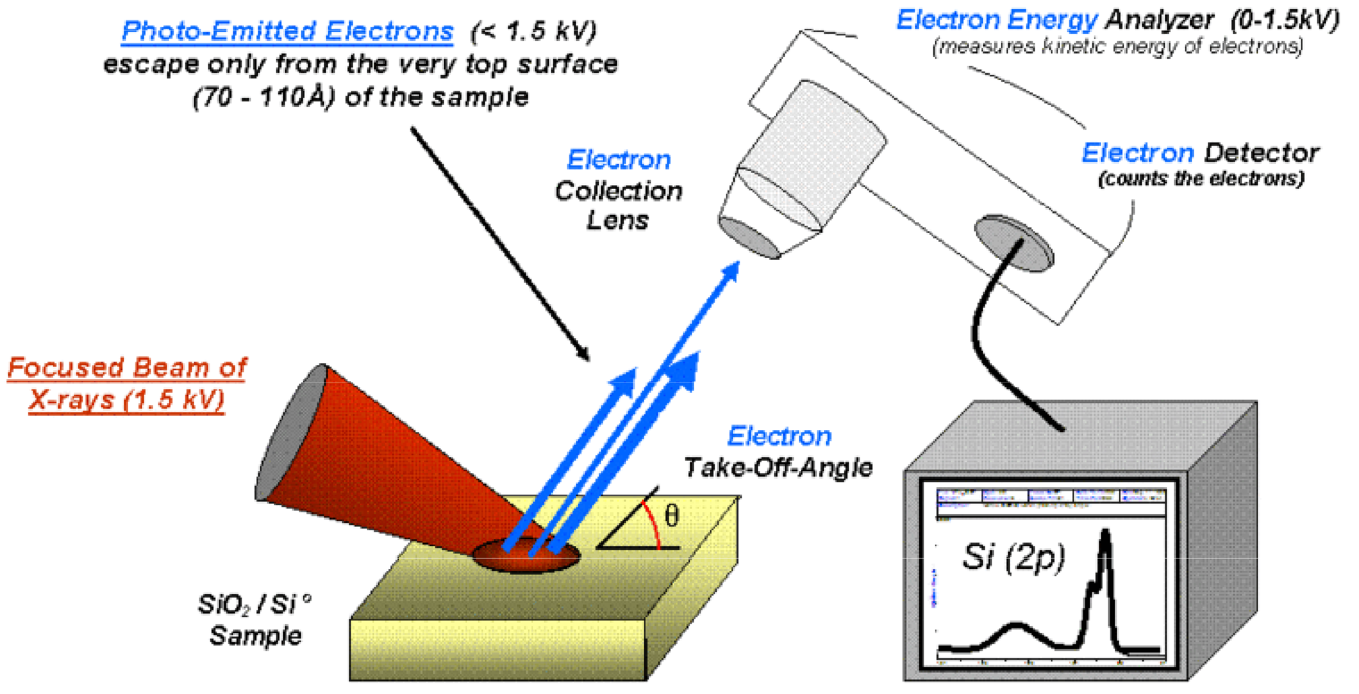


Fig. 3.25. BSE image of a copper–arsenic alloy. The alpha phase is composed of regular grains, containing 3% As. Along the grain boundaries Cu_3As intermetallics, with an As concentration up to 28% (bright phase), are present next to copper sulphide inclusions (dark phase). Magnification 400 [67]. Reproduced with permission from TMS Publications.

Fotoelektronová spektroskopie (ESCA)



Samples are usually solid because XPS requires ultra-high vacuum ($<10^{-8}$ torr)

Si (2p) XPS signals from a Silicon Wafer

Ultrafialová (UPS)
Rentgenová (XPS)

Sledování kinetické energie fotoelektronů,
Ta závisí na energii molekulového orbitalu.

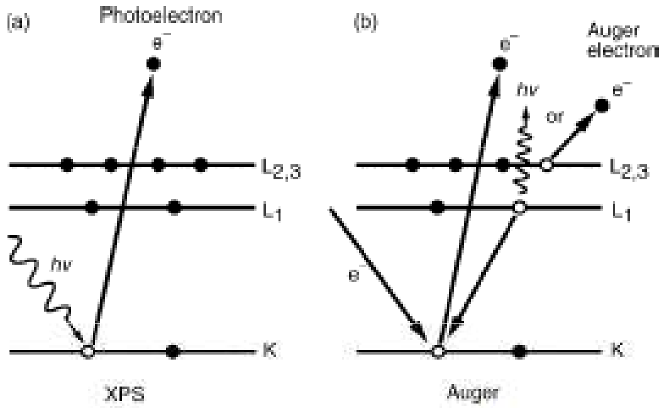


Figure 7.1 Emission processes of characteristic electrons: (a) a 1s photoelectron; and (b) a $KL_1L_{2,3}$ Auger electron

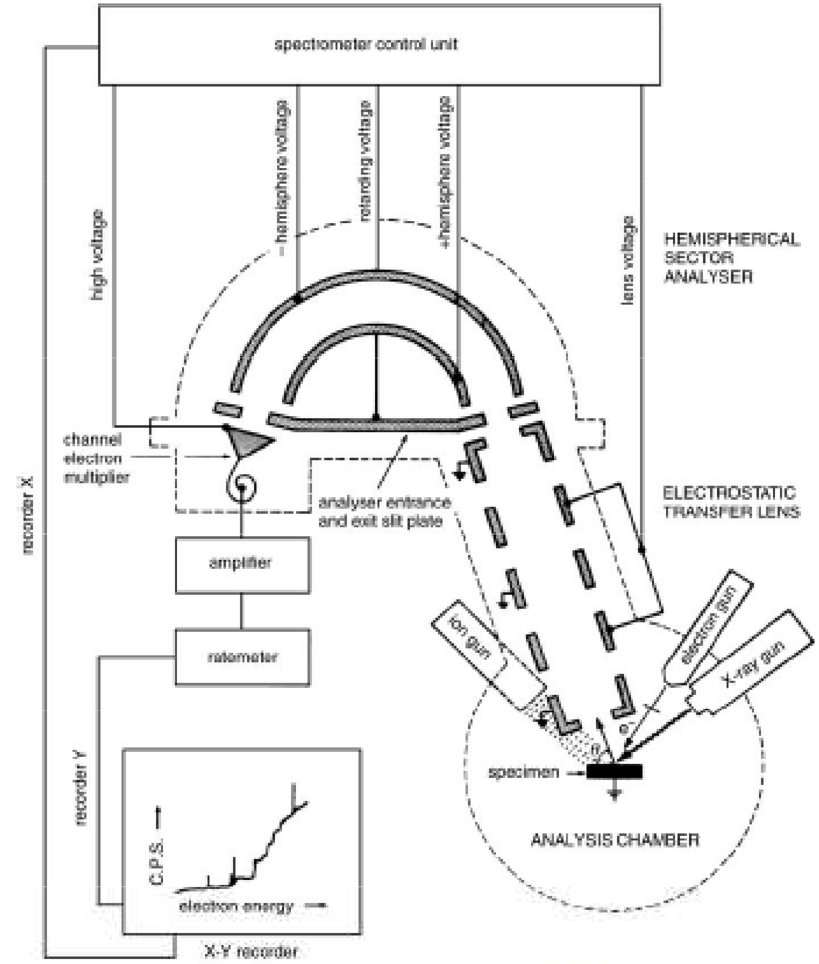


Figure 7.5 Structure of an electron spectrometer. (Reproduced with permission from J.F. Watts, *An Introduction to Surface Analysis by Electron Spectroscopy*, Oxford University Press, Oxford. © 1990 Royal Microscopy Society.)

PIXE a PIGE

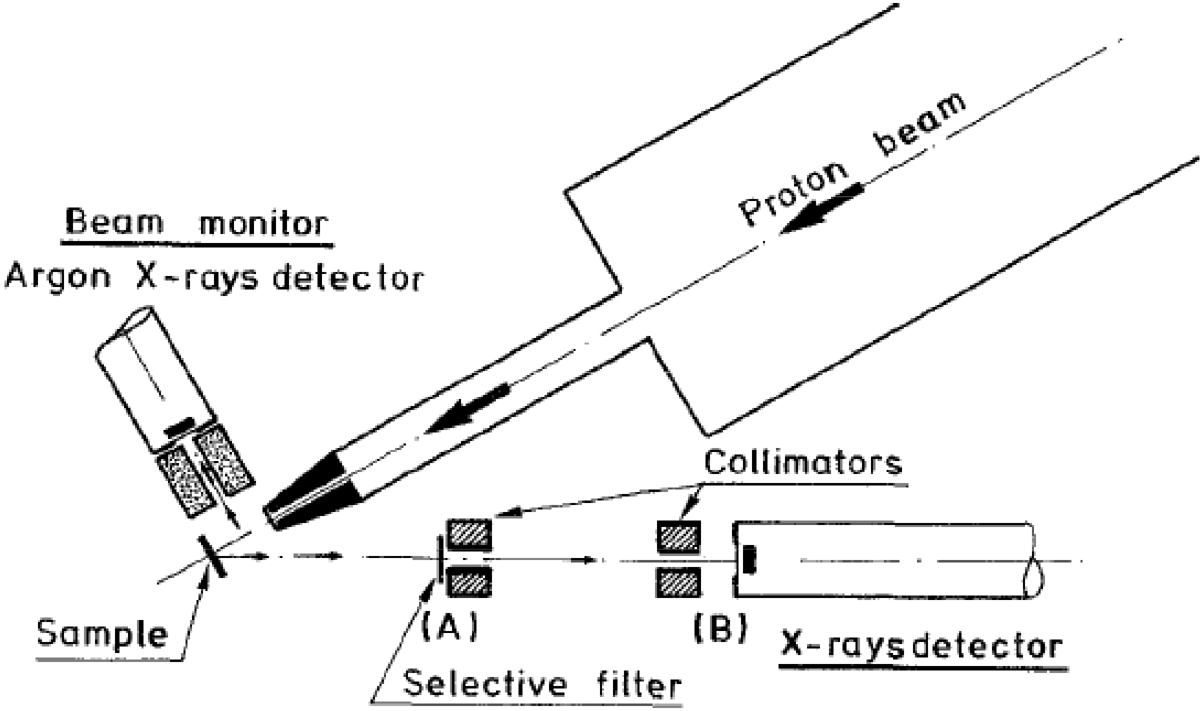


Fig. 11.1. The non-vacuum PIXE set-up used at LARN.

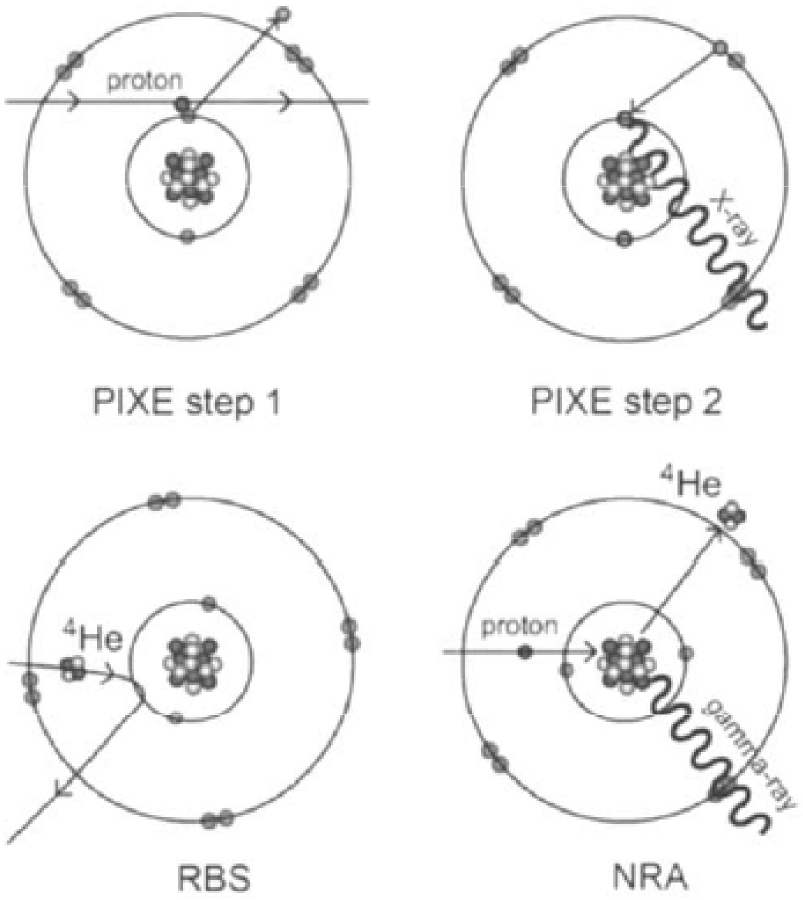
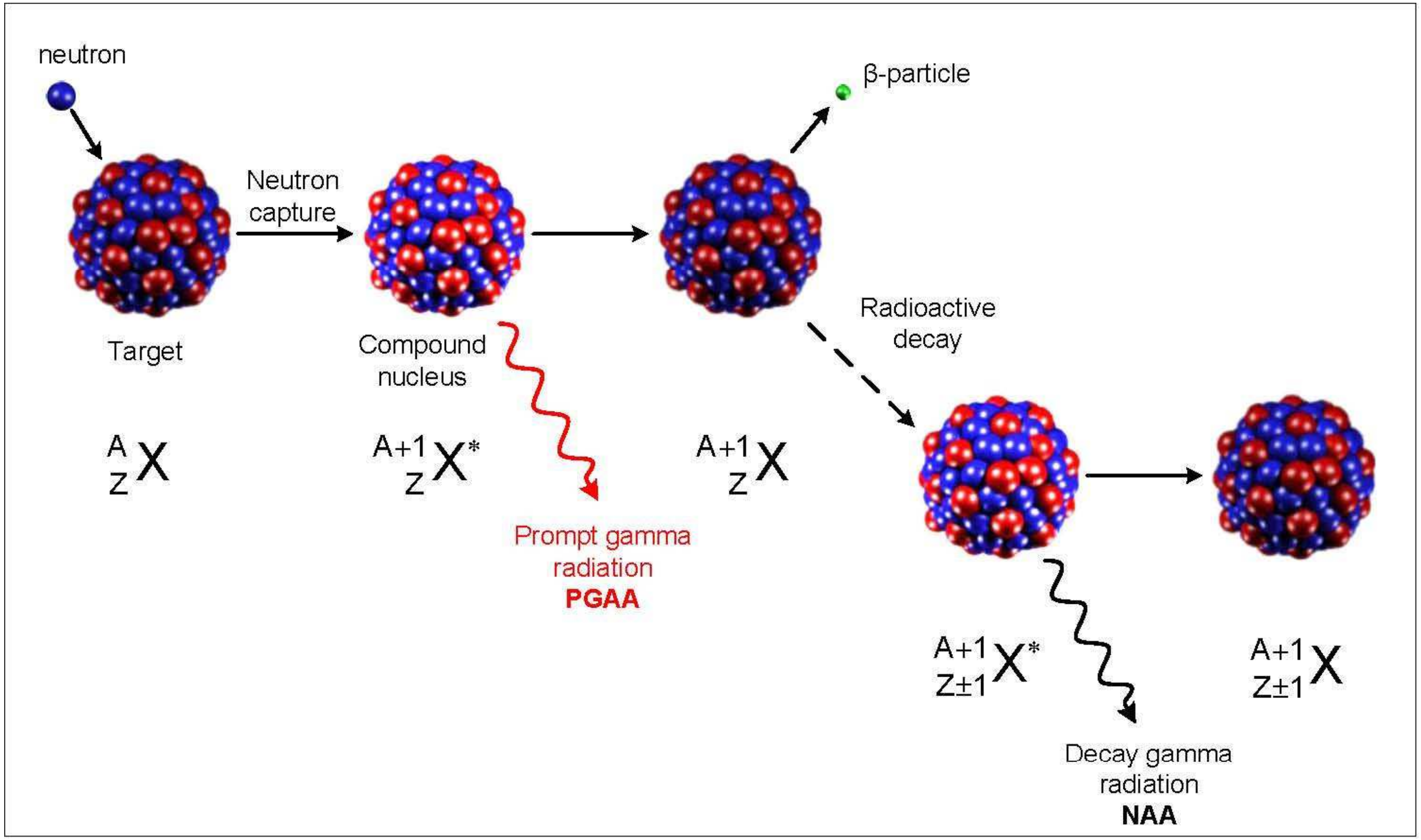


Fig. 5.1. Physical principles of IBA techniques. Particle-induced X-ray emission (PIXE) is a two-step process: an inner-shell electron of the target atom is expelled by the impinging ion, then follows an electronic rearrangement accompanied by X-ray emission. Rutherford backscattering spectrometry (RBS) relies on a purely elastic process based upon the electrostatic repulsion between positively charged projectile and target nuclei. NRA occurs when the projectile and the target nuclei come close enough to undergo a nuclear reaction with emission of characteristic photons or charged particles.

Neutronová aktivační analýza



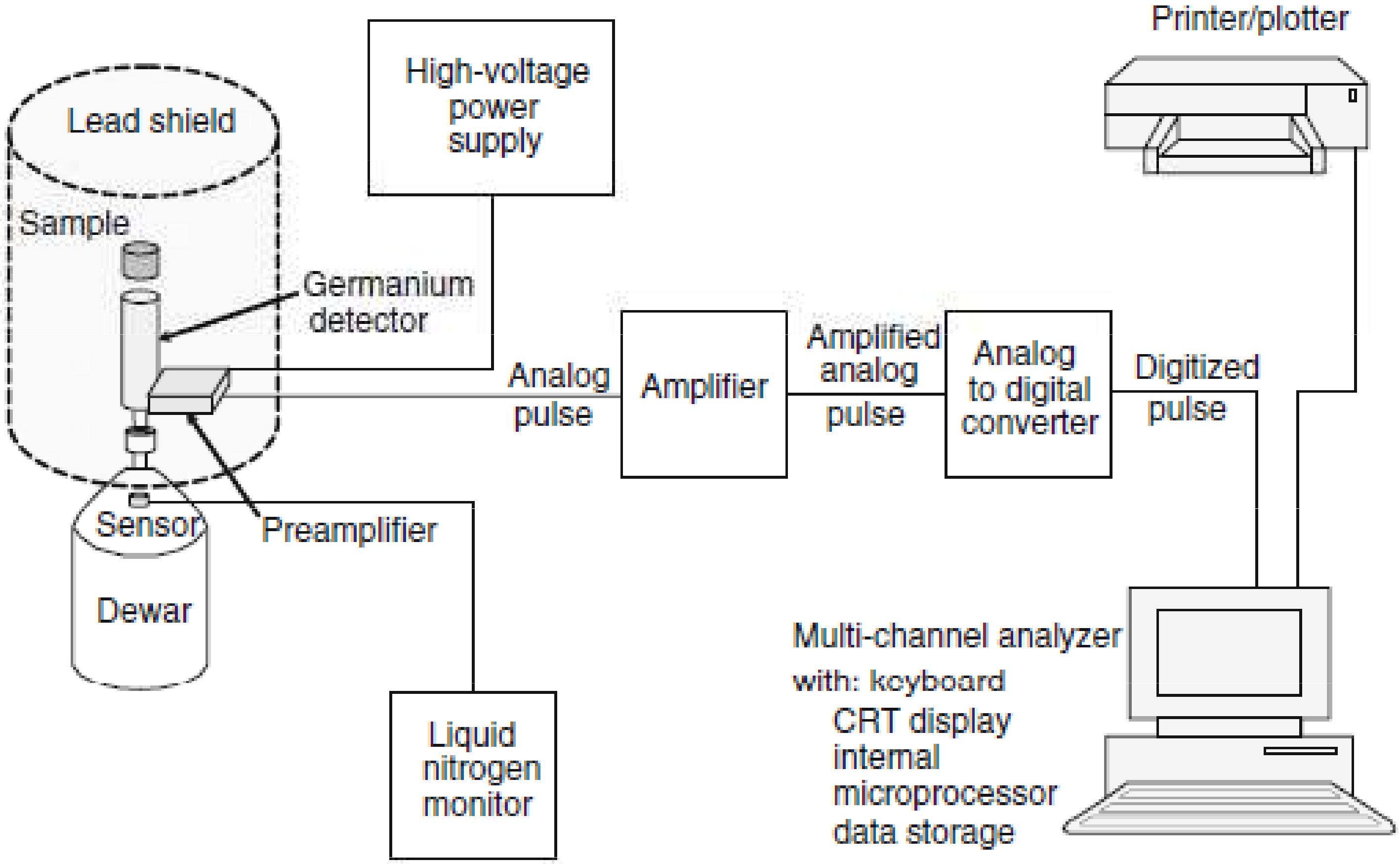


Fig. 32.1 Germanium–lithium detector (From SKOOG. Principles of Instrumental Analysis, 5E. © 1998 Brooks/Cole, a part of Cengage Learning, Inc. Reproduced by permission. www.cengage.com/permissions)

Hmotnostní spektrometrie

Izotopy

[a] The detection limits are based on a 98% confidence level (3 standard deviations).

[b] Identifying a single part per trillion of an element in a solution is analogous to locating a single white raisin in a house (2,700 sq. ft) full of regular raisins.

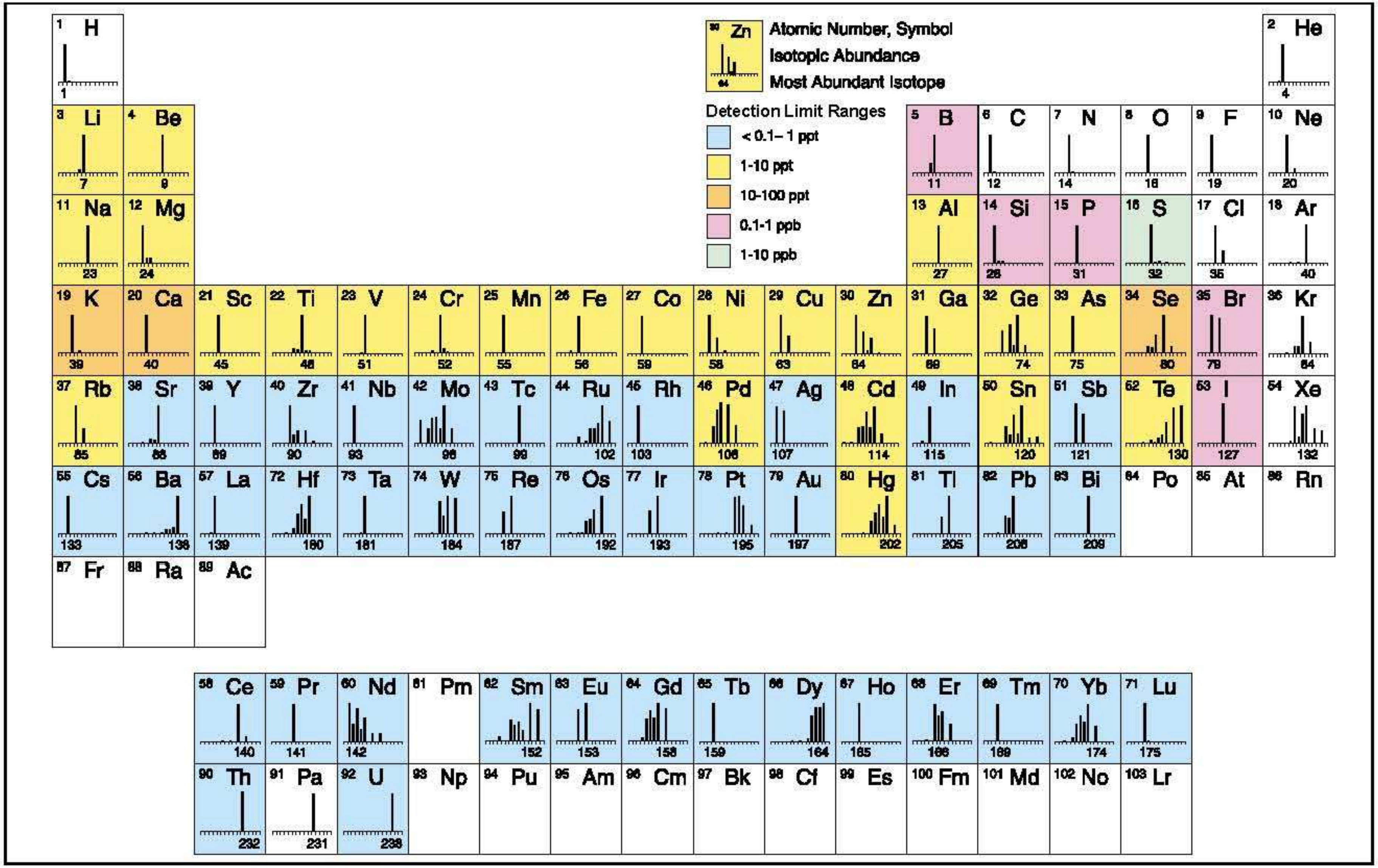


Figure 1. Elements determined by ICP-MS and approximate detection capability.

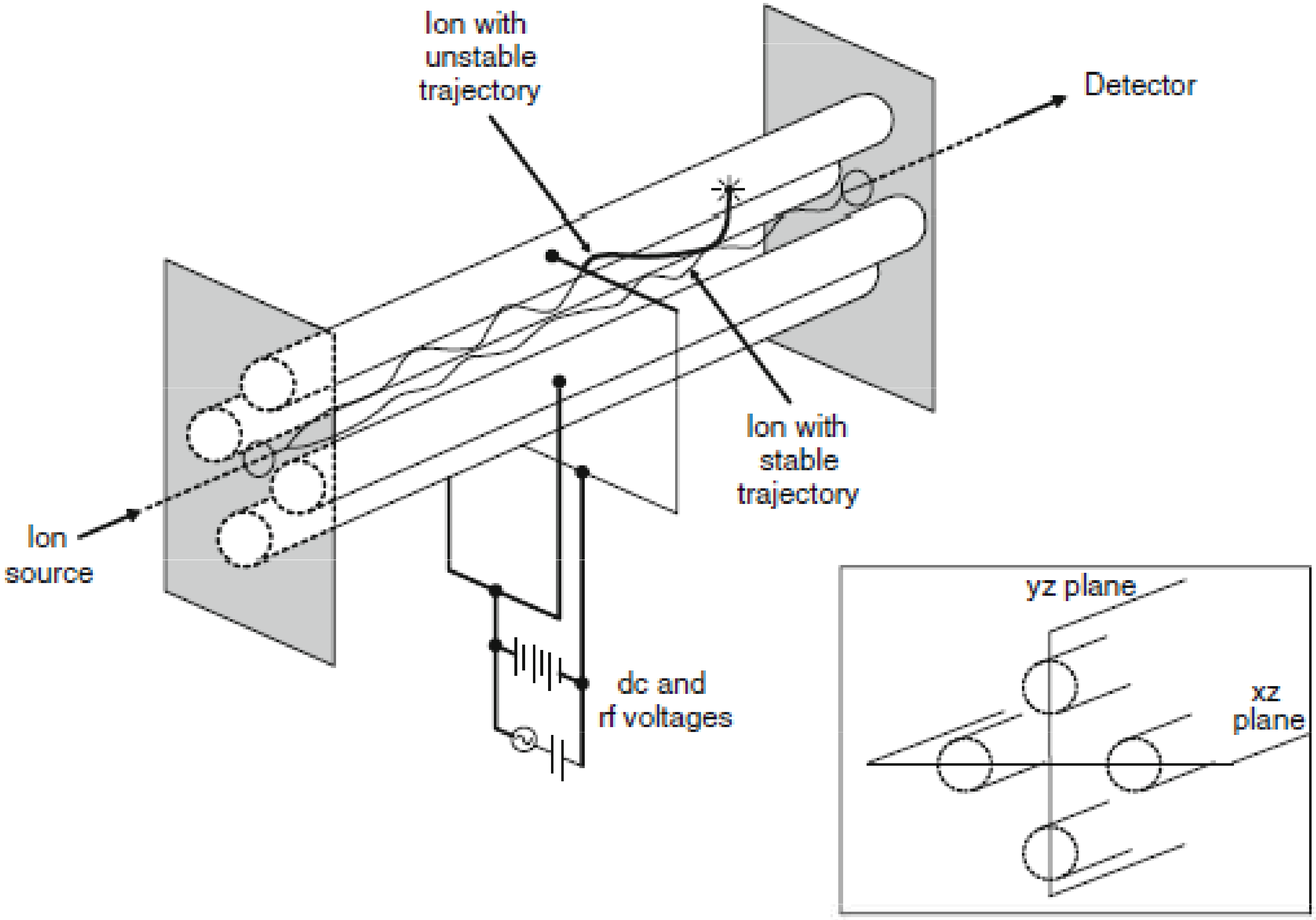


Fig. 31.5 Quadrupole mass spectrometer with inset of *xz* and *yz* planes (From SKOOG. Principles of Instrumental Analysis, 5E. © 1998 Brooks/Cole, a part of Cengage Learning, Inc. Reproduced by permission. www.cengage.com/permissions)

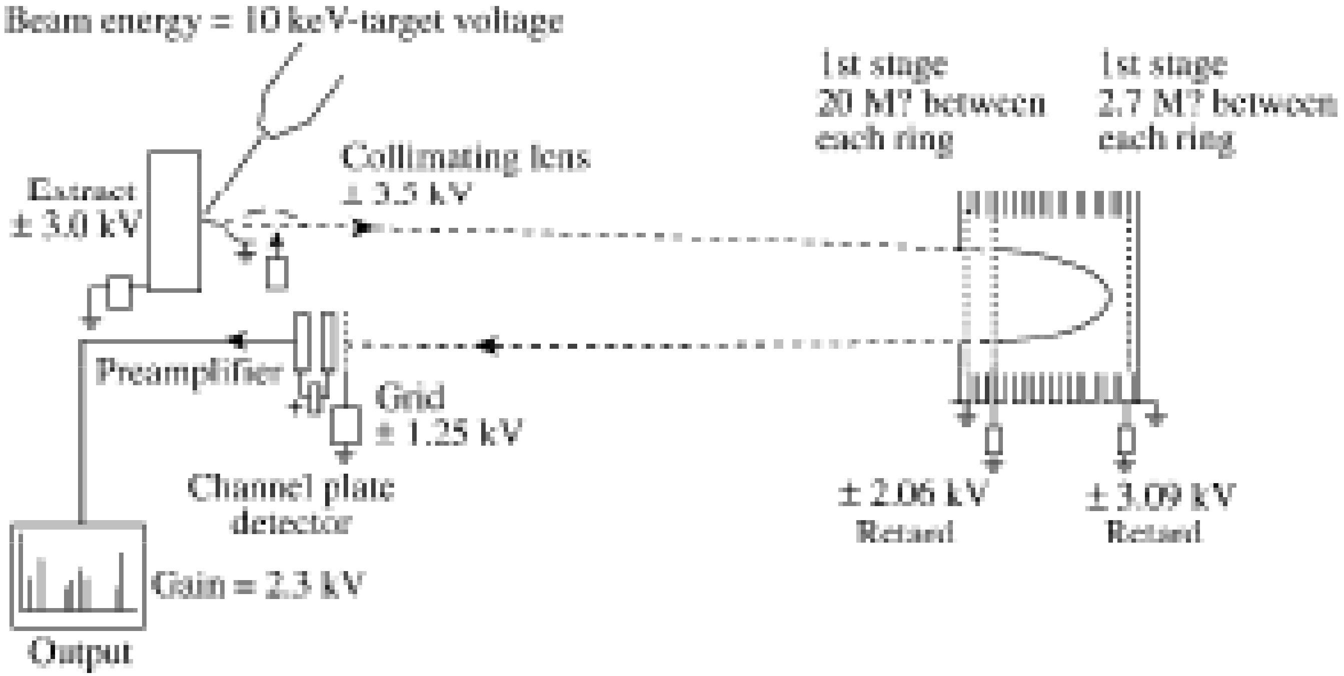
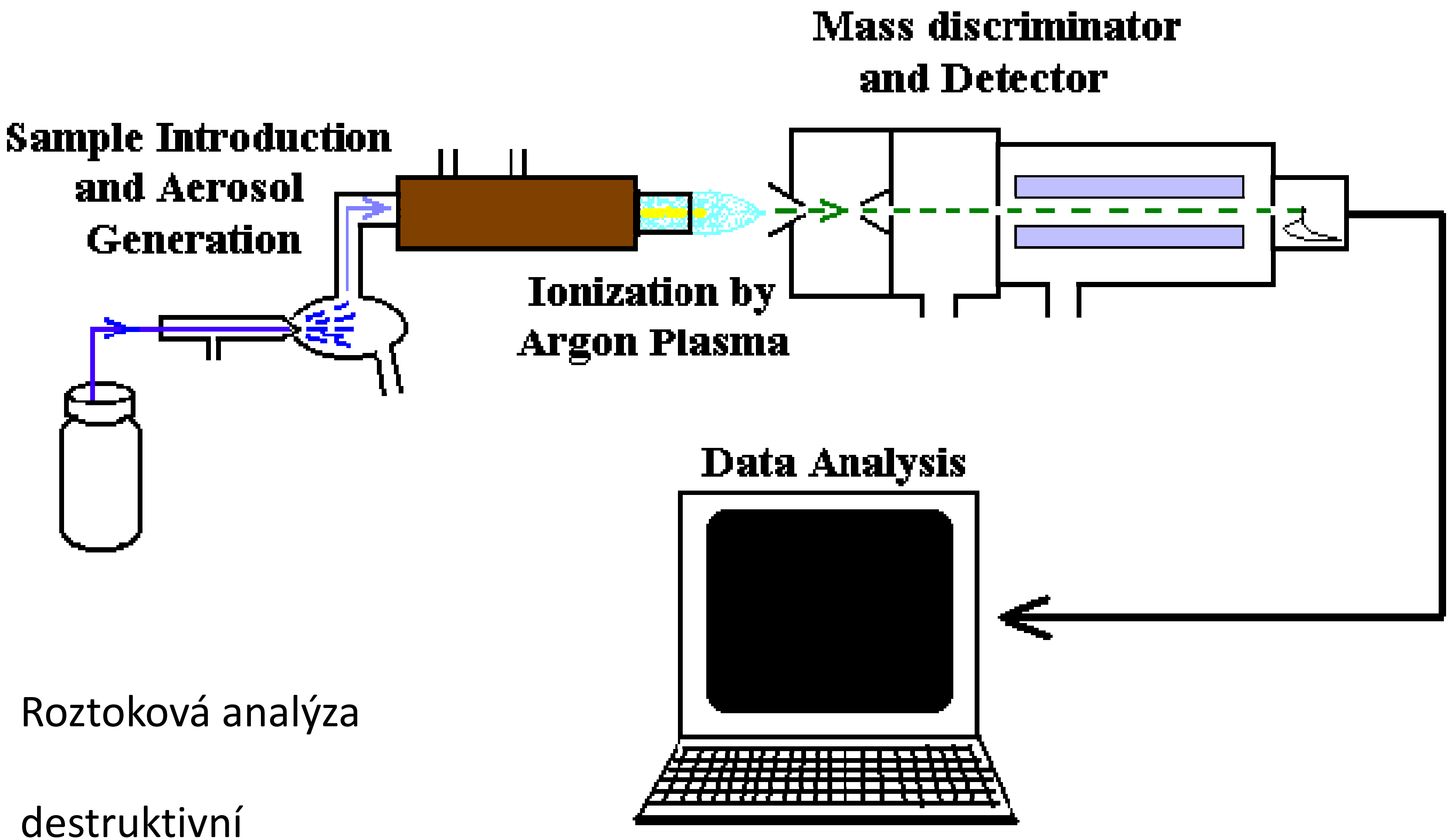


Figure 8.12 Time-of-flight mass analyzer. The secondary ion beam is reflected by a mirror to correct the flight time of ions with identical mz^{-1}. (Reproduced with permission from J.C. Vickerman, *Surface Analysis: The Principal Techniques*, John Wiley & Sons Ltd, Chichester. © 1997 J.C. Vickerman.)

Hmotnostní spektrometrie s indukčně vázaným plazmatem

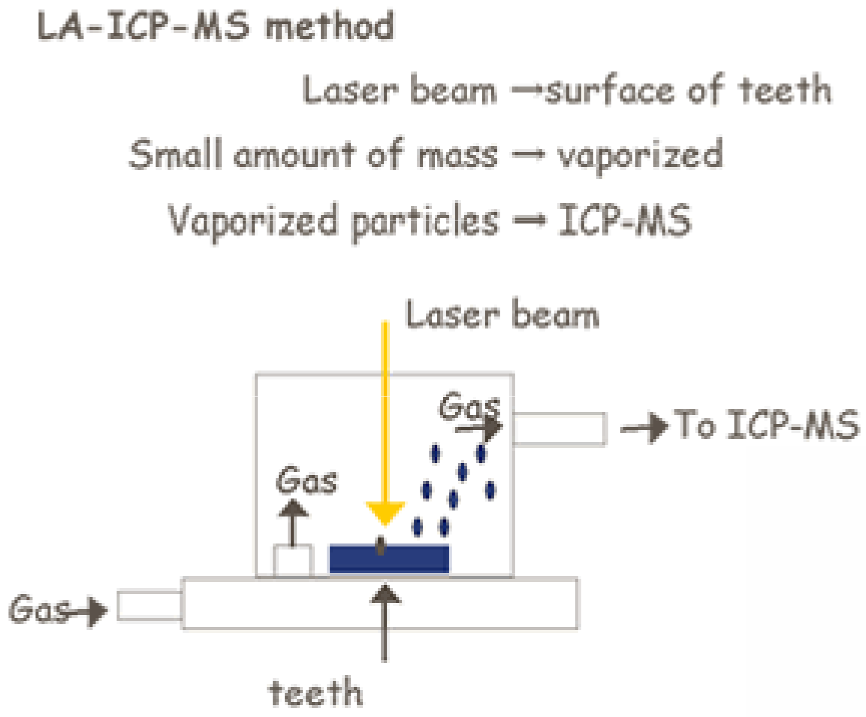


Roztoková analýza

destruktivní

LA-ICP-MS

Pevné látky vzorkovány laserovou ablací



LA-ICP-MS method

Laser beam →surface of teeth

Small amount of mass → vaporized

Vaporized particles → ICP-MS

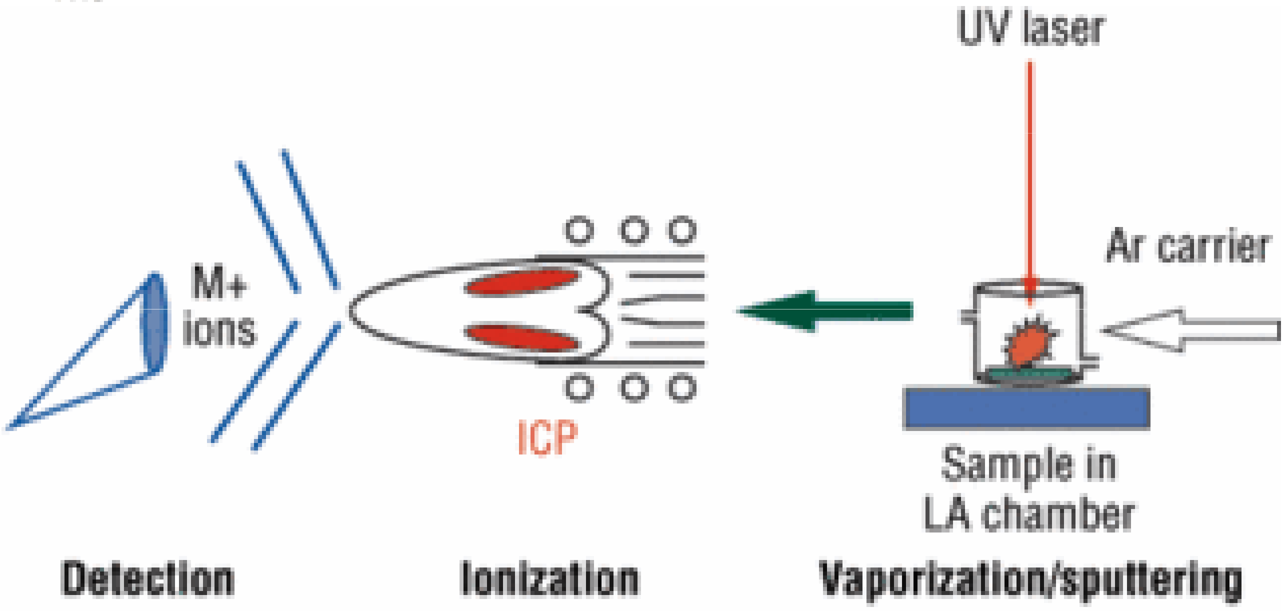
Nd:YAG

Excimer

Kvadrupólový

TOF

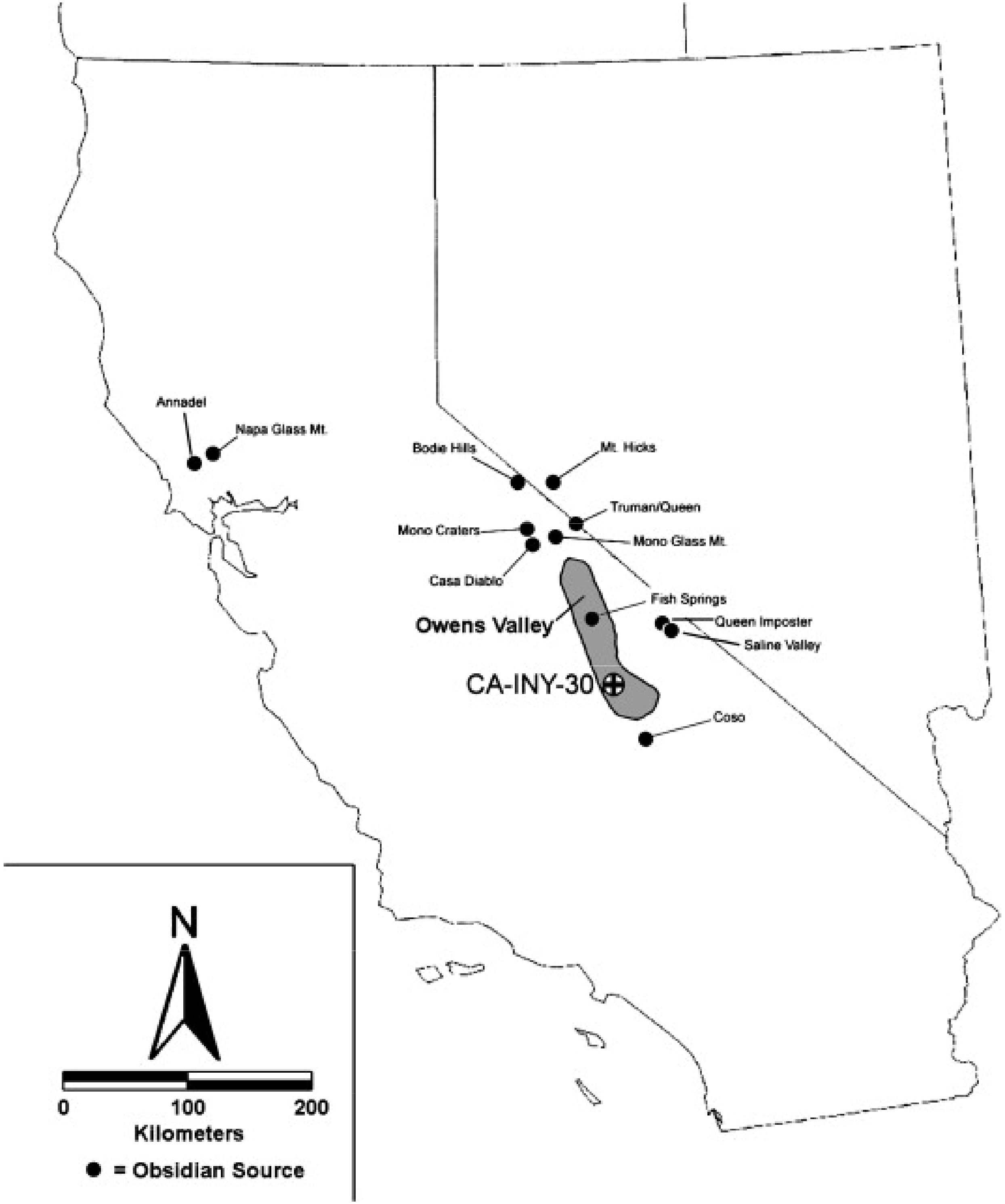
Sektorový (MC)

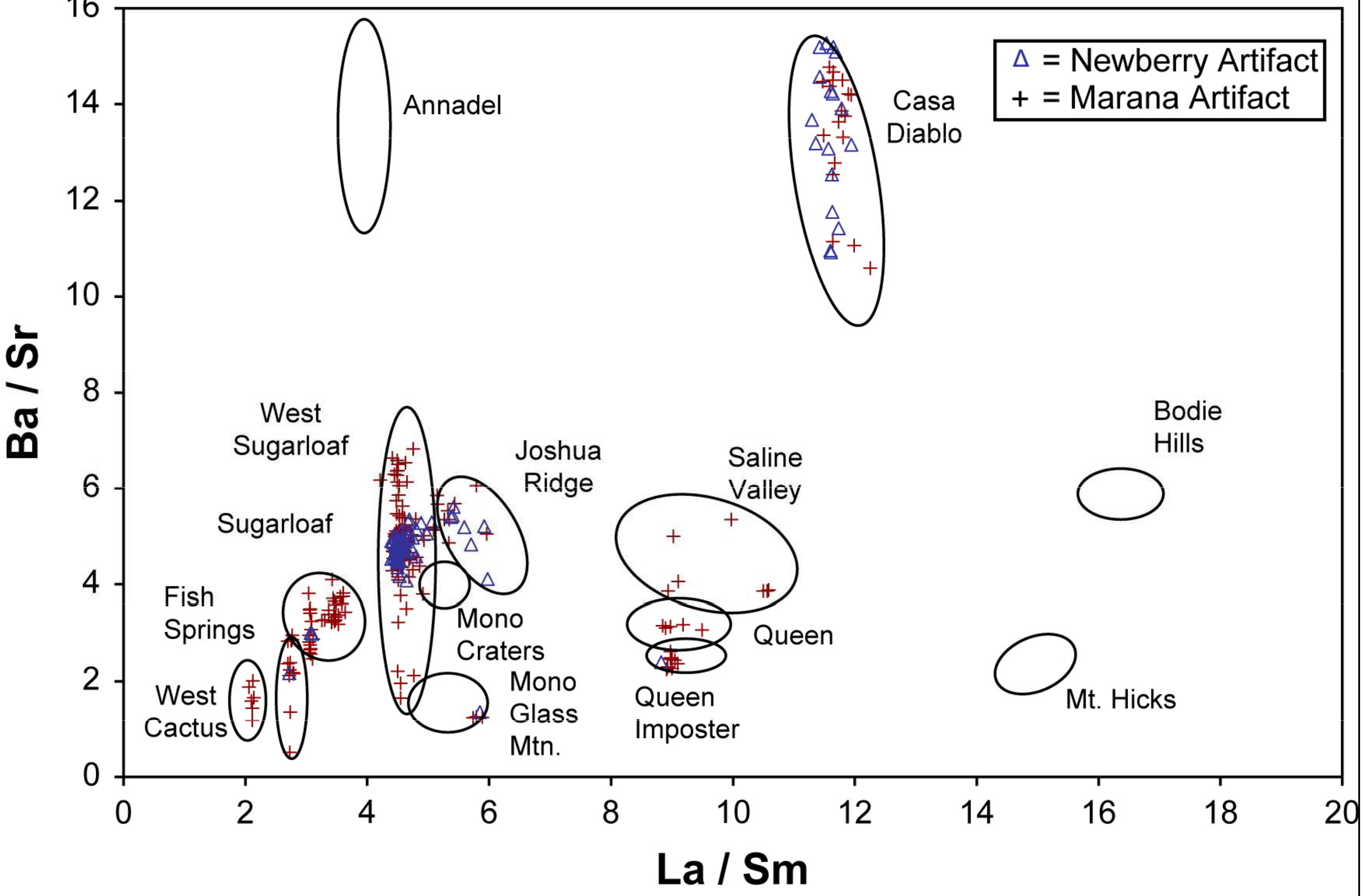


Semidestruktivní metoda

Provenience obsidiánu

Owens Valley,
vých. Kalifornie




Ba / Sr
La / Sm
0
2
4
6
8
10
12
14
16
0
2
4
6
8
10
12
14
16
18
20
Δ = Newberry Artifact
+ = Marana Artifact
Annadel
Casa Diablo
West Sugarloaf
Sugarloaf
Joshua Ridge
Saline Valley
Bodie Hills
Fish Springs
West Cactus
Mono Craters
Mono Glass Mtn.
Queen
Queen Imposter
Mt. Hicks

Malba na keramice

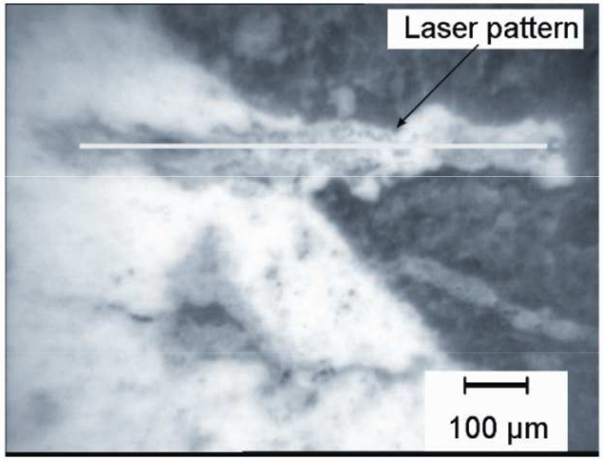


Fig. 5. Post-ablation photograph showing line pattern which has been ablated across the slip-decoration boundary of a Mimbres pottery sherd

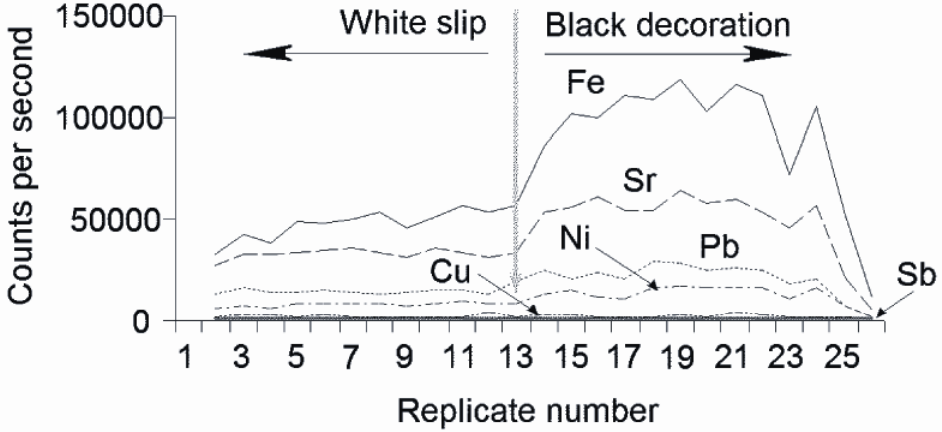
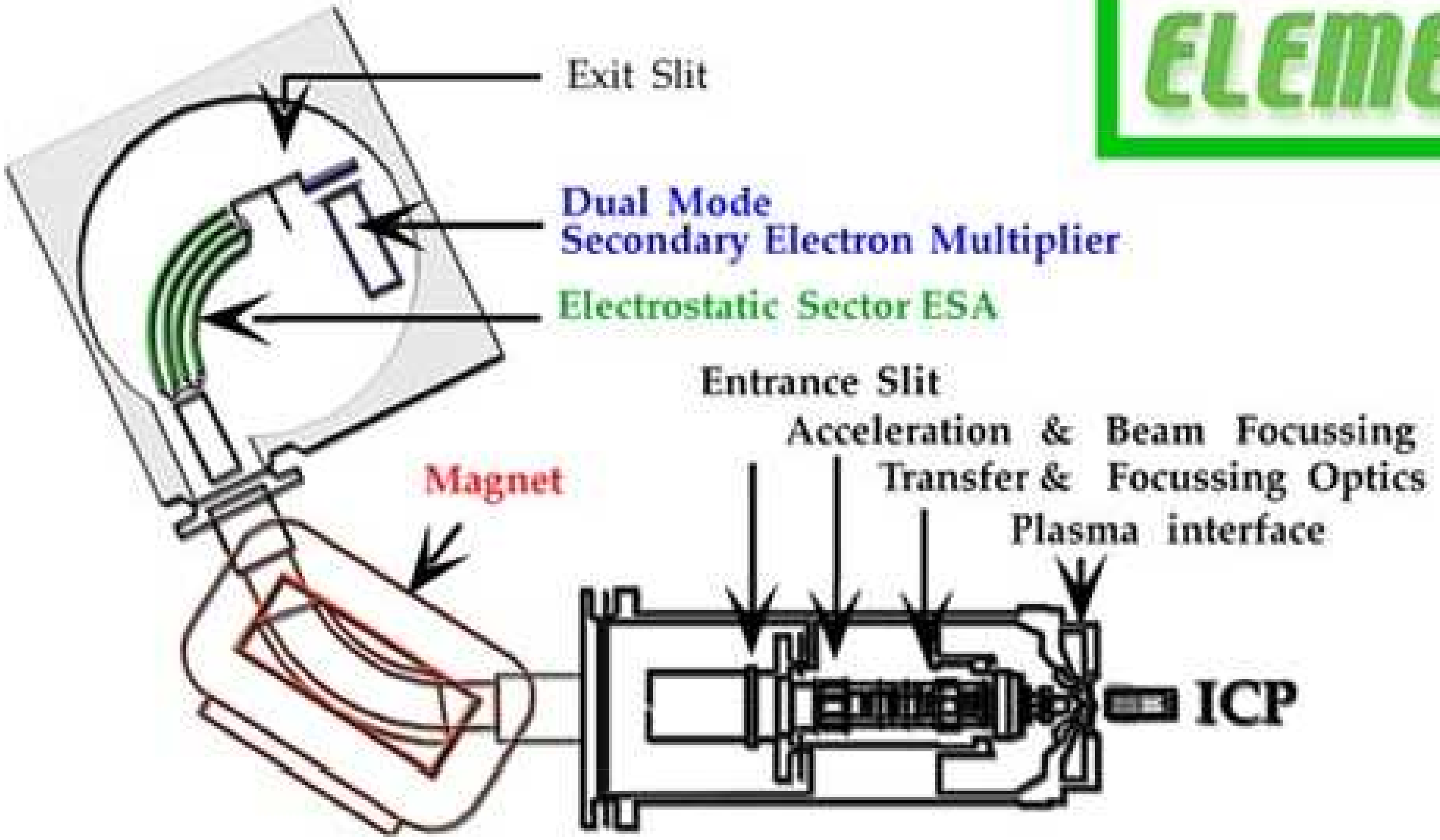


Fig. 6. Elemental scan showing replicate analyses over time as laser pattern is ablated across white slip to black decoration border on a Mimbres pottery sherd

MC LA-ICP-MS

Analýza izotopových poměrů

ELEMENT 2



Analýza izotopových poměrů

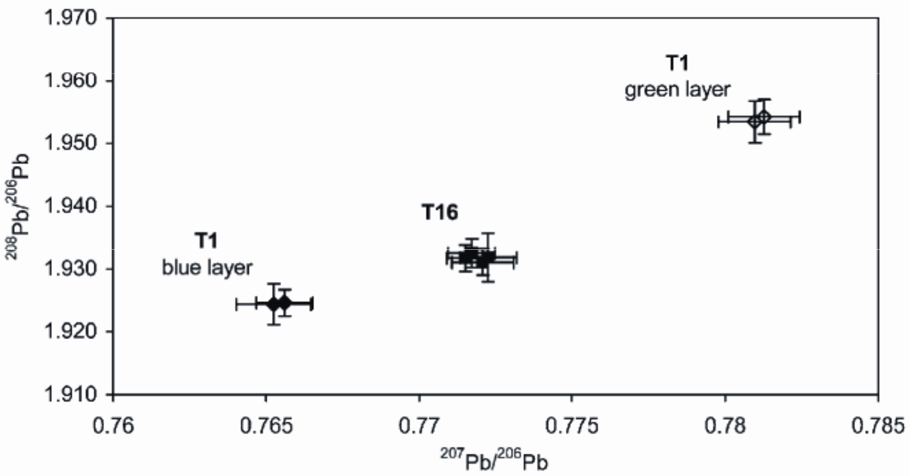


Fig. 2 Lead isotope ratio distribution of the two iridescent Art Nouveau glasses. Glass T1 consists of two layers of approximately equal thickness (coloured green and blue, respectively). Error bars represent total combined uncertainties ($k = 1$).

Sklo „Art nouveau“

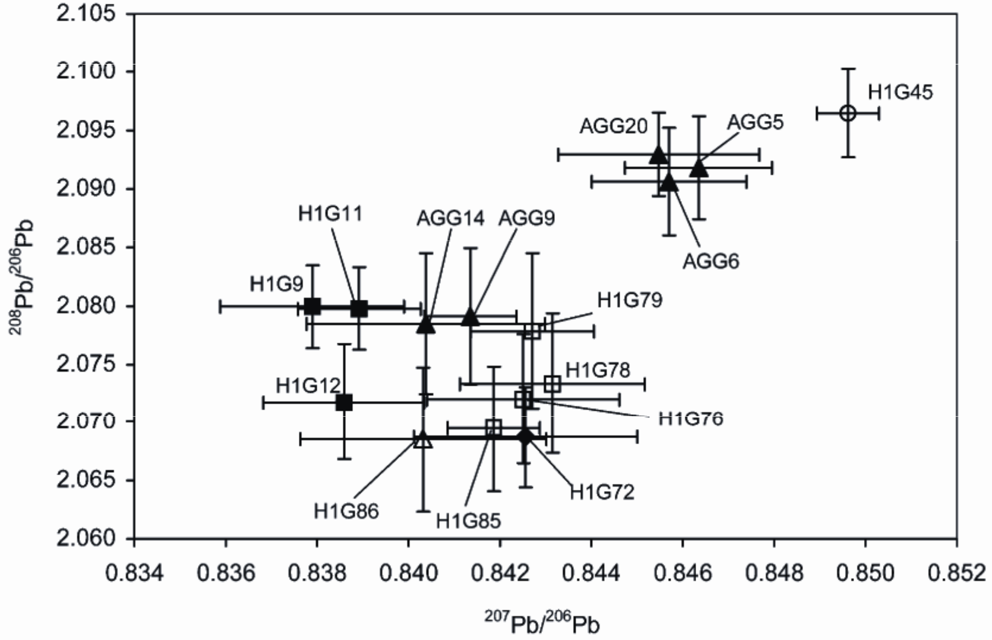
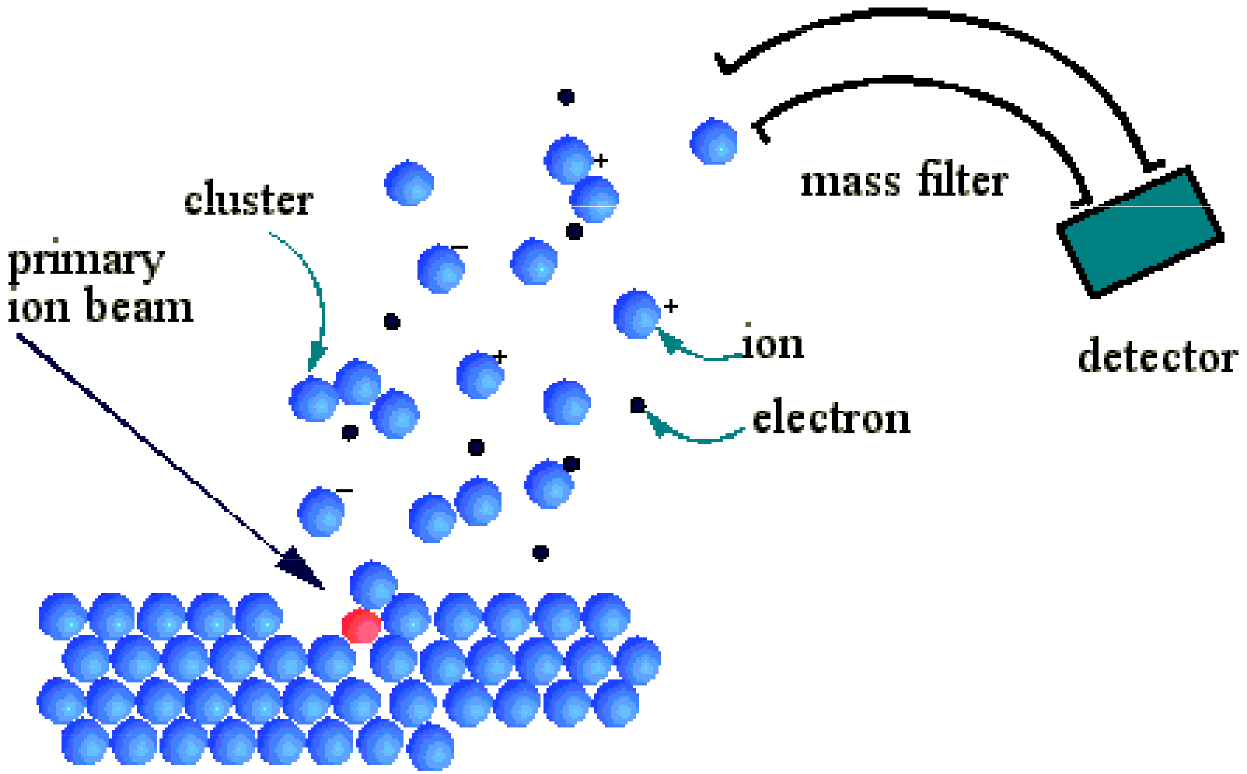


Fig. 3 Lead isotope ratios of Ephesos glass samples after external mass bias correction *via* the NIST SRM 610 glass reference material. Error bars represent total combined uncertainties ($k = 1$). For sample description see Table 1.

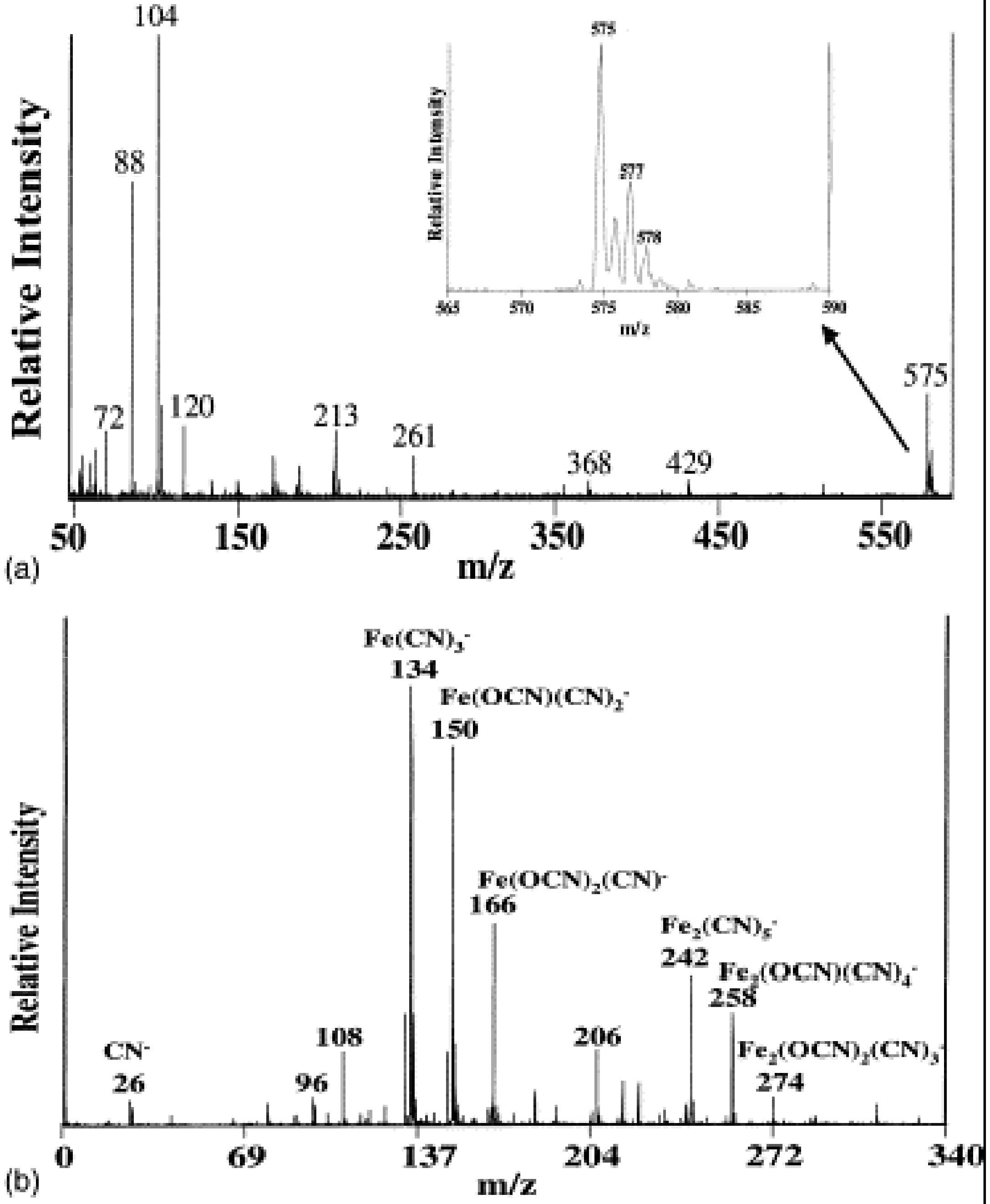
LDI - TOF

Desorpce a ionizace laserem



Analýza pigmentů

Pruská modř



m/z	Assignment	Fe oxidation states
108	$Fe(CN)_2^-$	+1
124	$Fe(OCN)(CN)^-$	+1
134	$Fe(CN)_3^-$	+2
150	$Fe(OCN)(CN)_2^-$	+2
166	$Fe(OCN)_2(CN)^-$	+2
182	$Fe(OCN)_3^-$	+2
190	$Fe_2(CN)_3^-$	(+1, +1) or (0, +2)
206	$Fe_2(OCN)(CN)_2^-$	(+1, +1) or (0, +2)
216	$Fe_2(CN)_4^-$	(+1, +2) or (0, +3)
222	$Fe_2(OCN)_2(CN)^-$	(+1, +1) or (0, +2)
242	$Fe_2(CN)_5^-$	(+2, +2) or (+1, +3)
258	$Fe_2(OCN)(CN)_4^-$	(+2, +2) or (+1, +3)
274	$Fe_2(OCN)_2(CN)_3^-$	(+2, +2) or (+1, +3)

Pigmenty v iluminovaných rukopisech

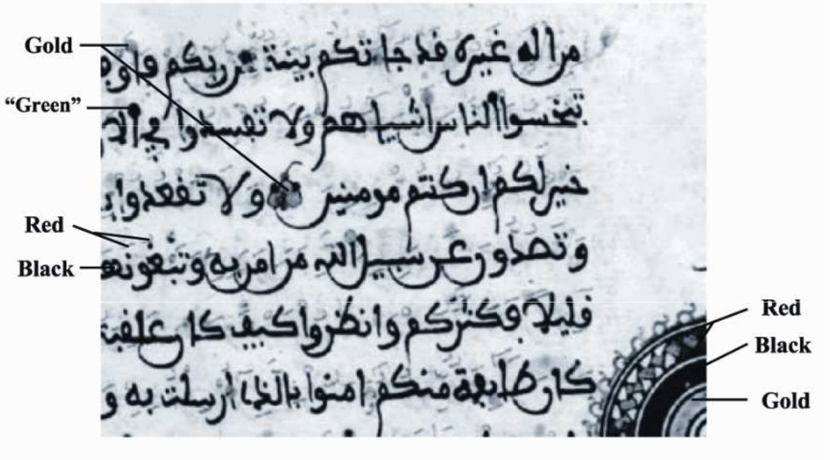


Figure 4 *A portion of the page of the Qur'an used in this study.*

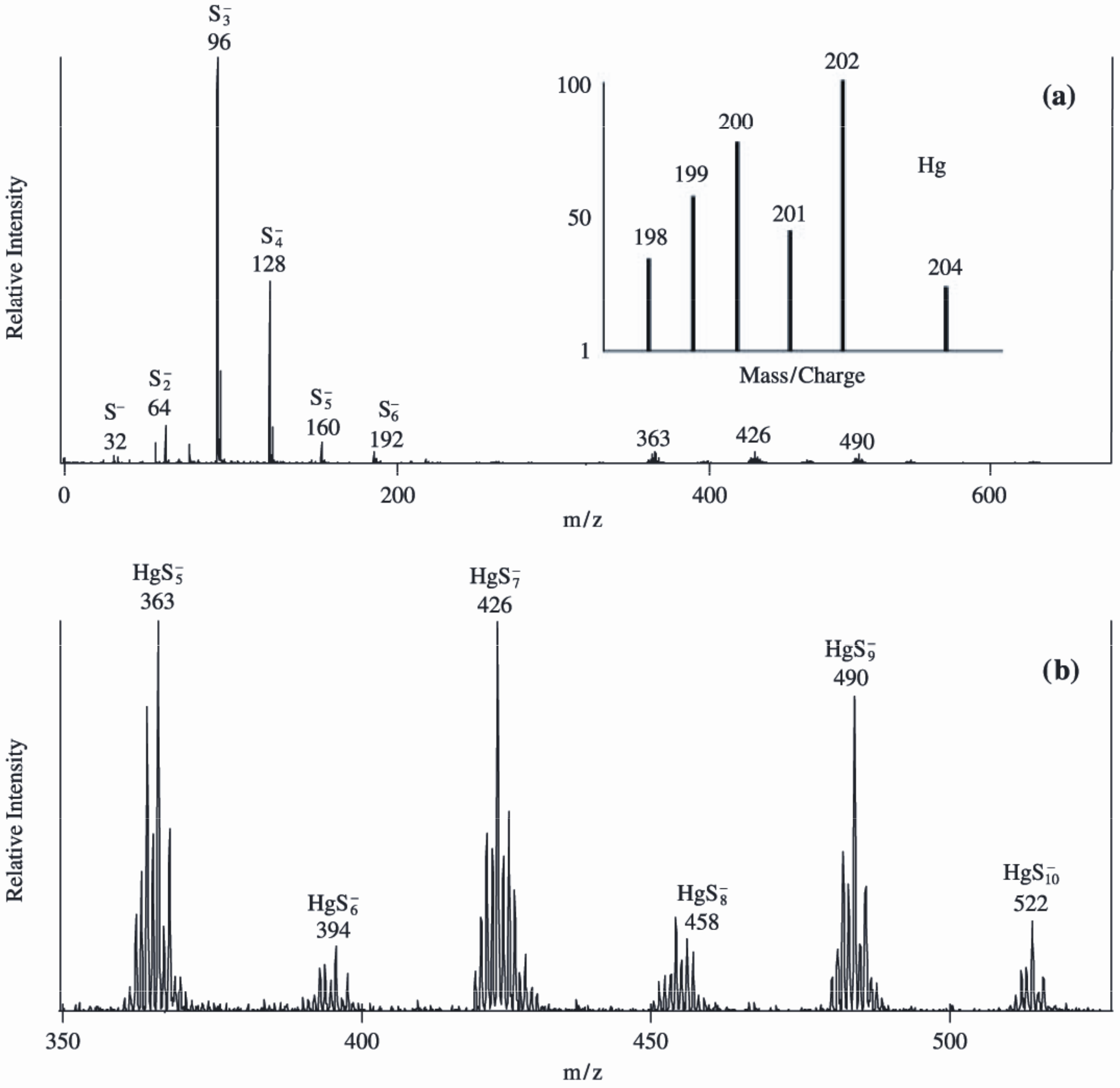


Figure 5 *(a) A negative ion LDMS spectrum of the red ink/dye region of the Qur'an sample. (b) An expanded view of the higher m/z portion of the spectrum.*

Akrylátové barvy

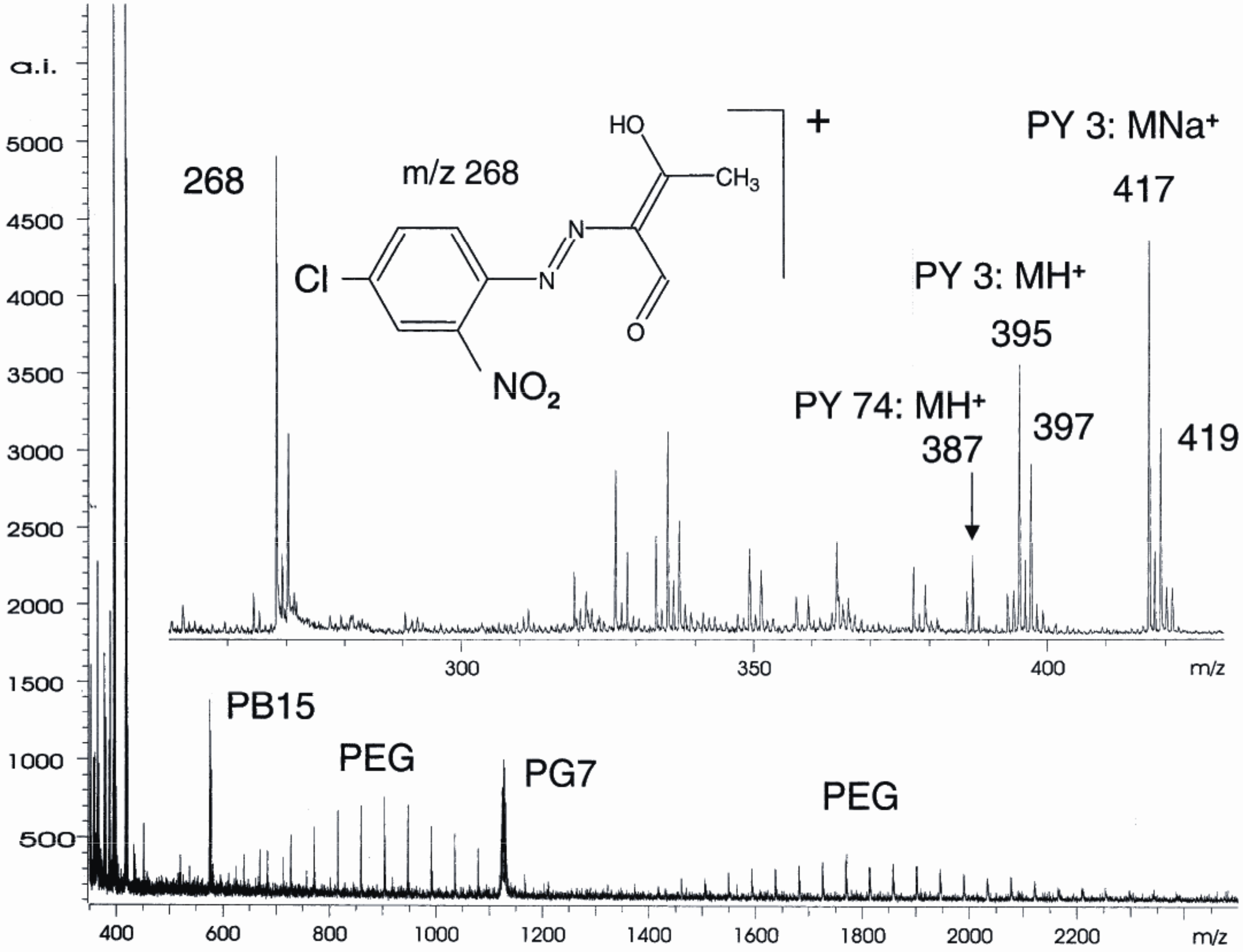
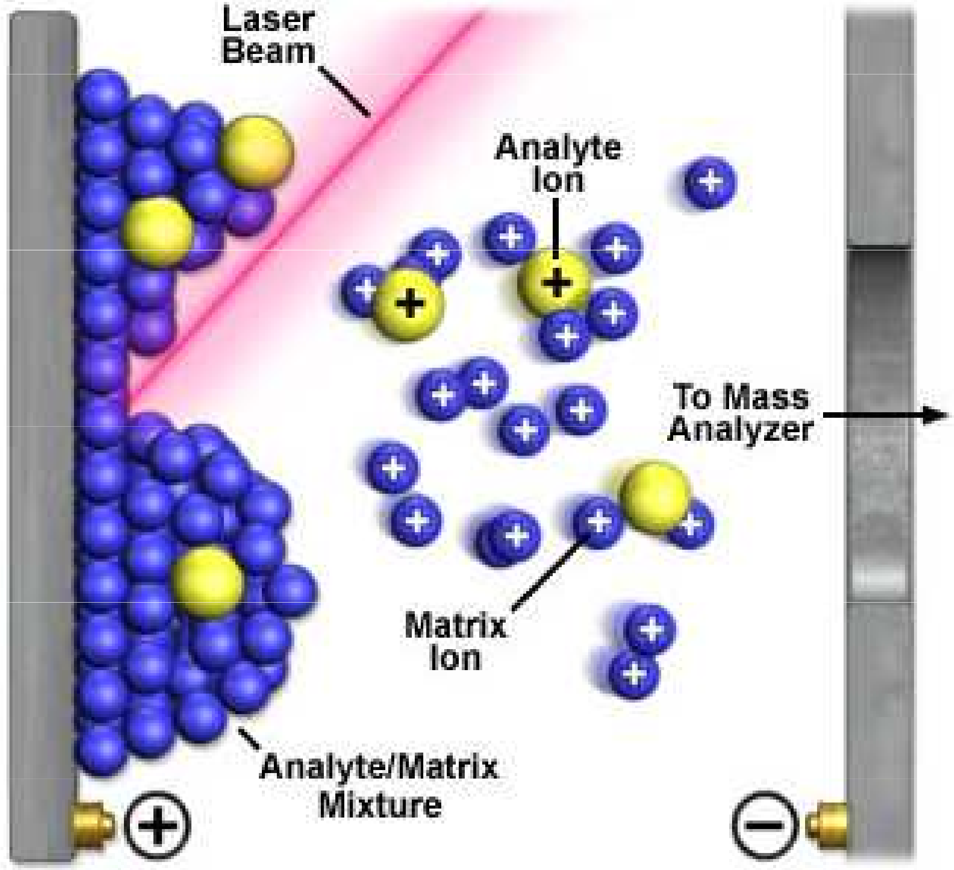


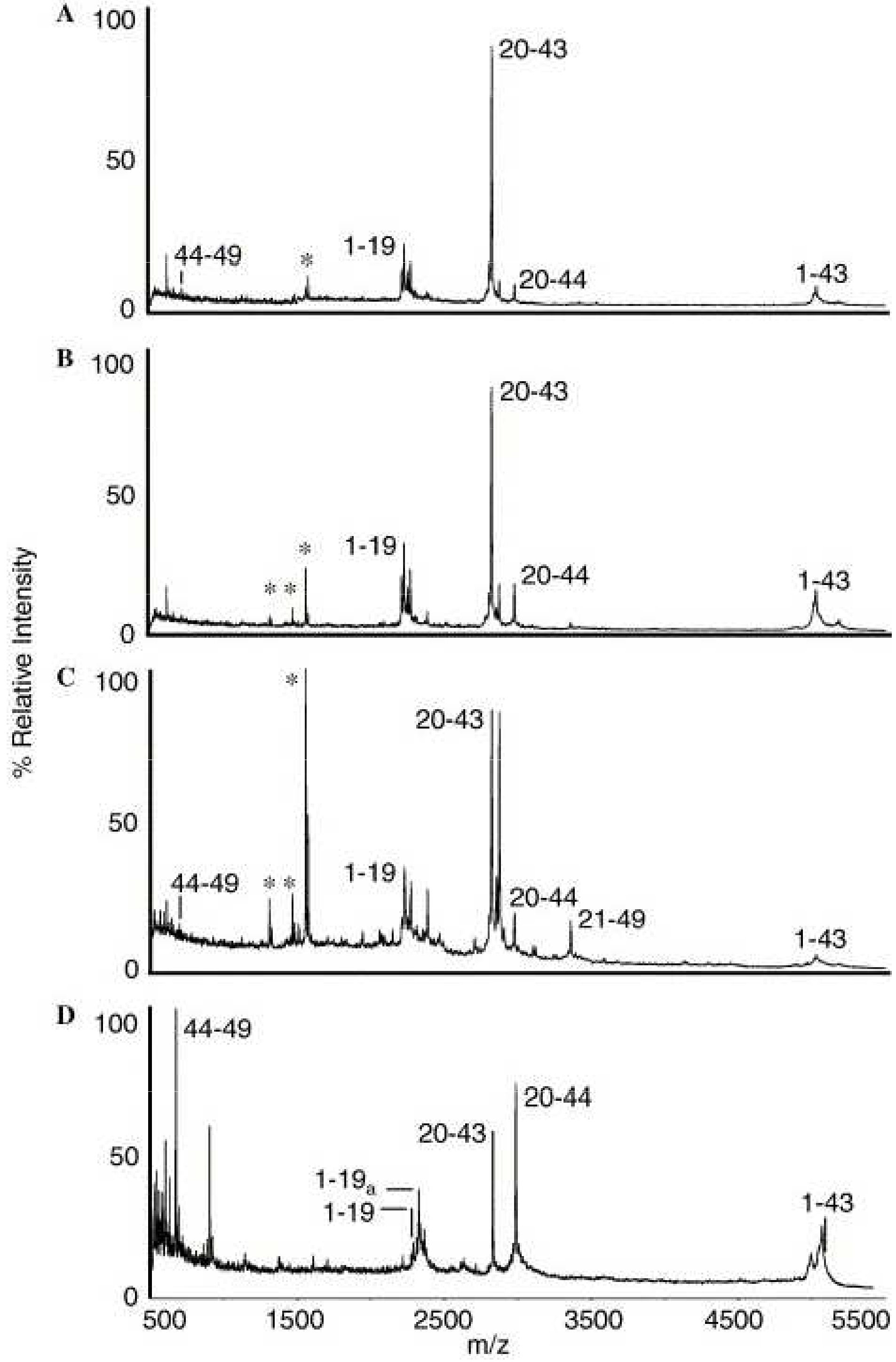
Fig. 11. Laser desorption ionisation mass spectrum of Winsor and Newton Finity ‘Permanent Green Light’ acrylic paint. The paint was painted on cellulose TLC plate surface and after drying analysed directly by LDI-TOFMS using a nitrogen laser at 337 nm.

MALDI-TOF

Desorpce a ionizace laserem za přítomnosti matrice



Analýza fosilních proteinů

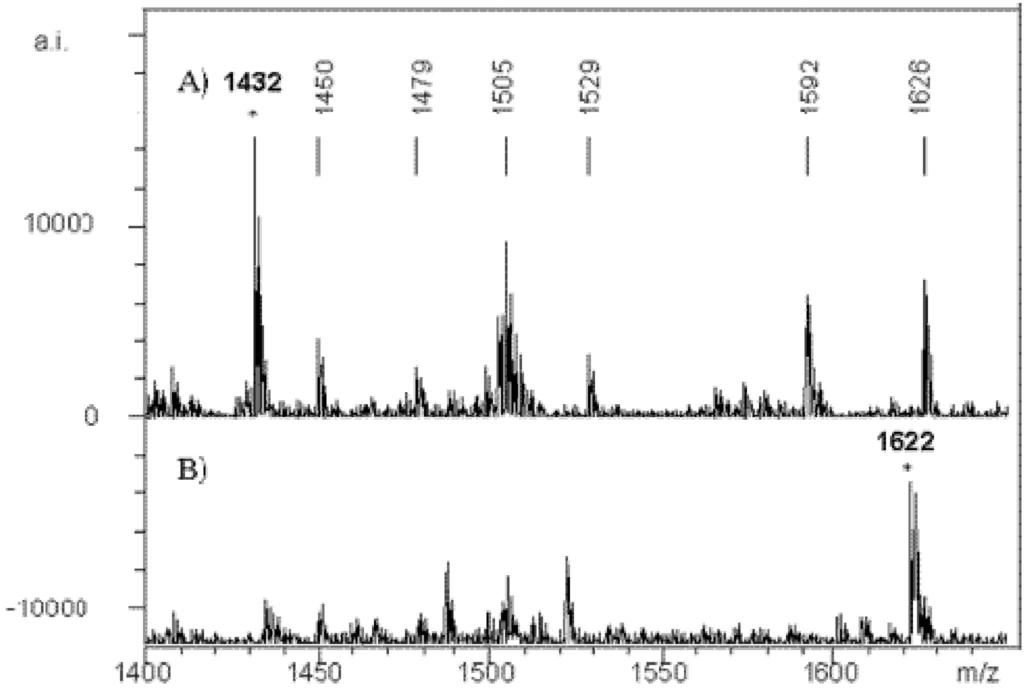


„Peptide mass fingerprint“ osteokalcinu rozloženého trypsinem:

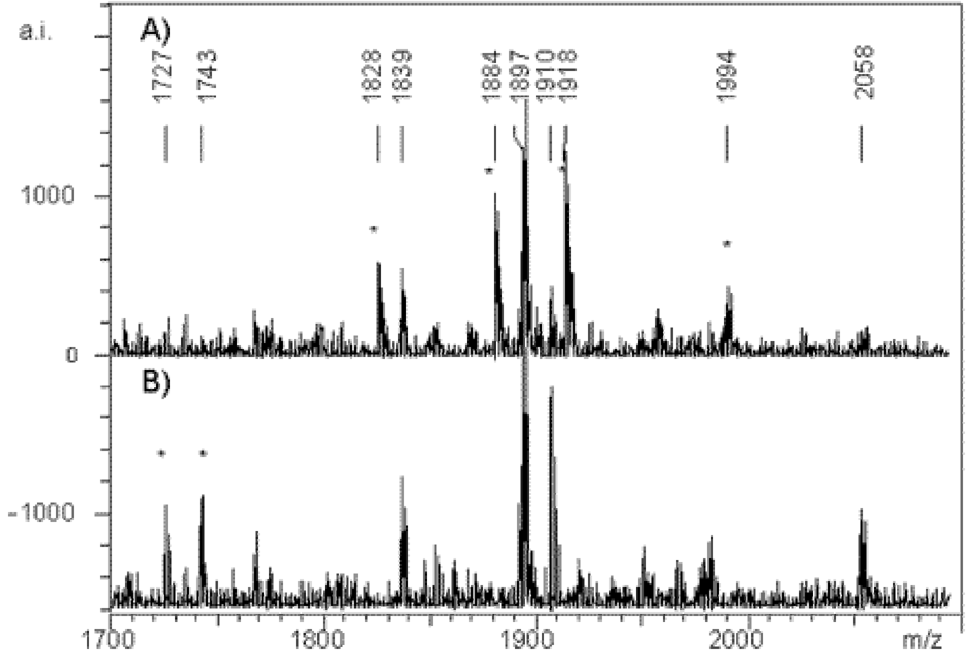
(A) extant horse
(B) zebra
(C) osel
(D) částečně čištěný osteokalcin z 42 000 let starých pozůstatků koně.

Identifikace keratinů

Hmotnostrní spektra 1400 to 1700 Da vlny jaka (A) a kašmírské kozy (B) (rozklad trypsinem).

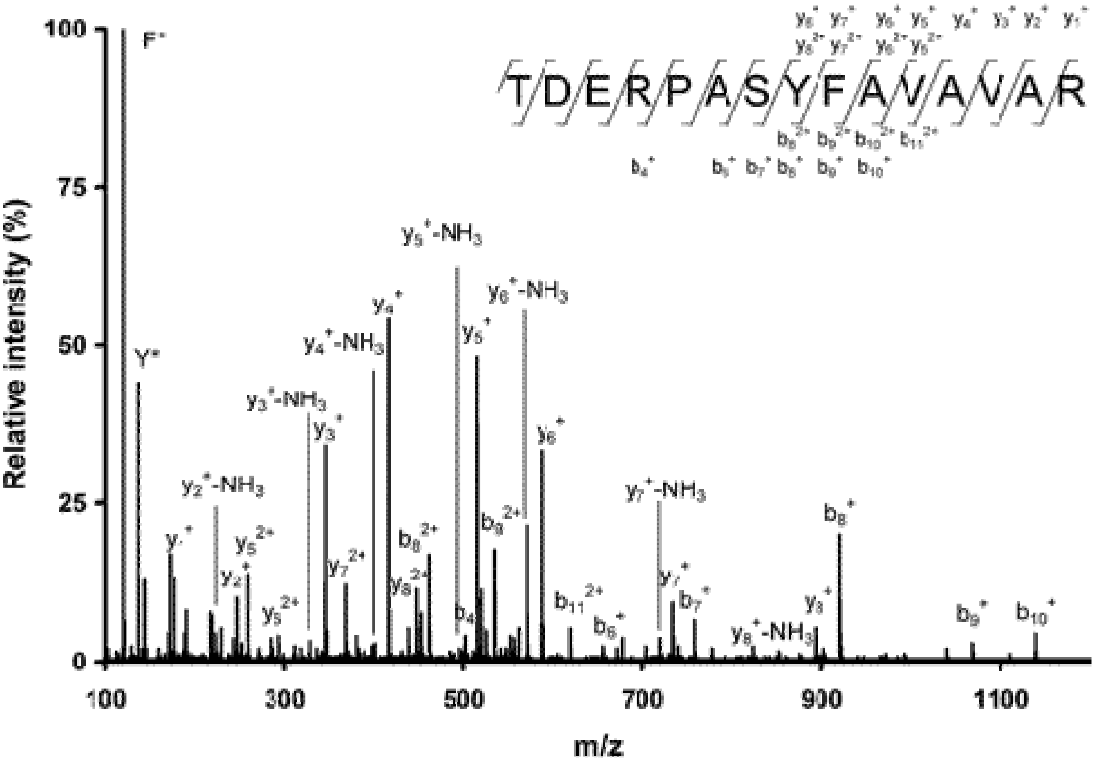


Hmotnostní spektra 1700 to 2100 Da peří husy (A) a kachny (B).



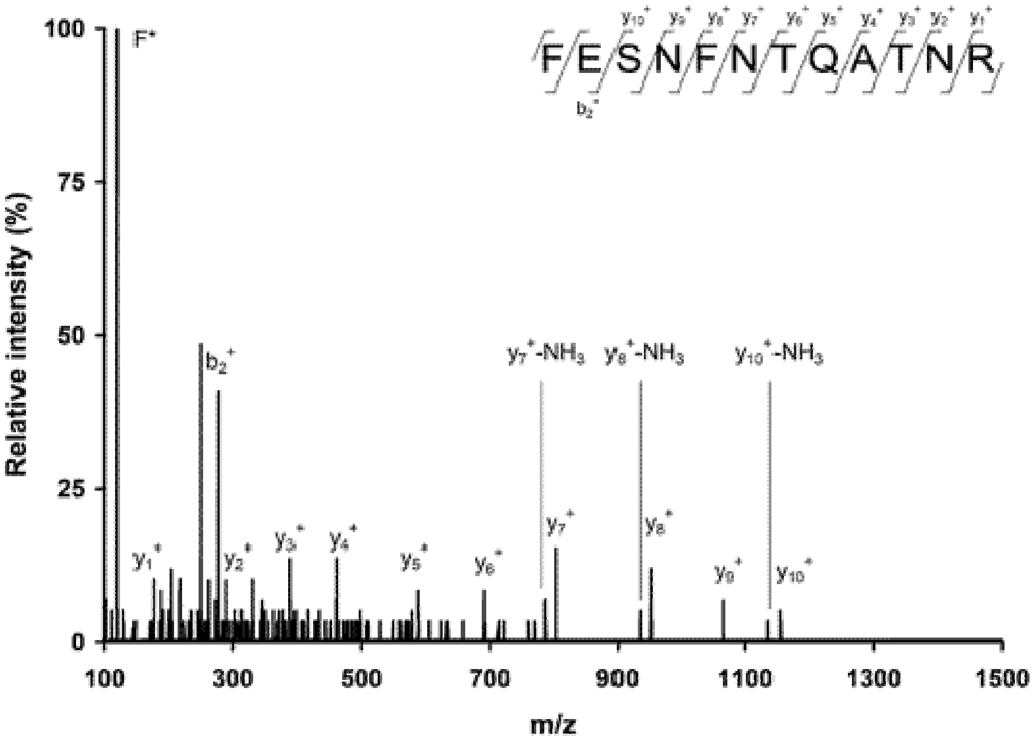
Identifikace organických pojiv v malbách

Triptych Benedetta Bonfigliho, Madona s dítětem, sv. Jan Křtitel.sv. Šebestián (XV. století).



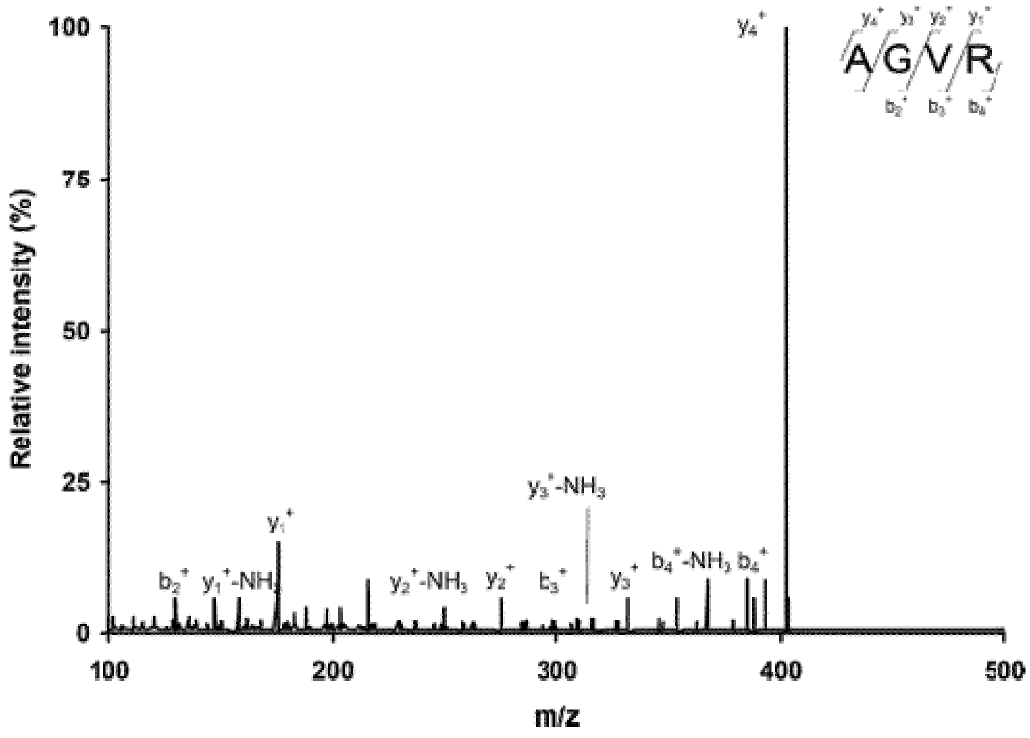
MS/MS spektrum trojnásobně nabitých iontů pro *m/z* 551.61, z hydrolyzovaného extraktu. Přítomny jsou **y** a **b** fragmenty peptidu ovotransferrinu 443-457 (TDERPASYFAVAVAR).

MS/MS spektrum dvojnásobně nabitých iontů *m/z* 714.82, z hydrolyzovaného extraktu z triptychu Benedetta Bonfigliho. Přítomny jsou fragmenty **y** and **b** fragmentu peptidu lysozymu 52-63 (FESNFNTQATNR).



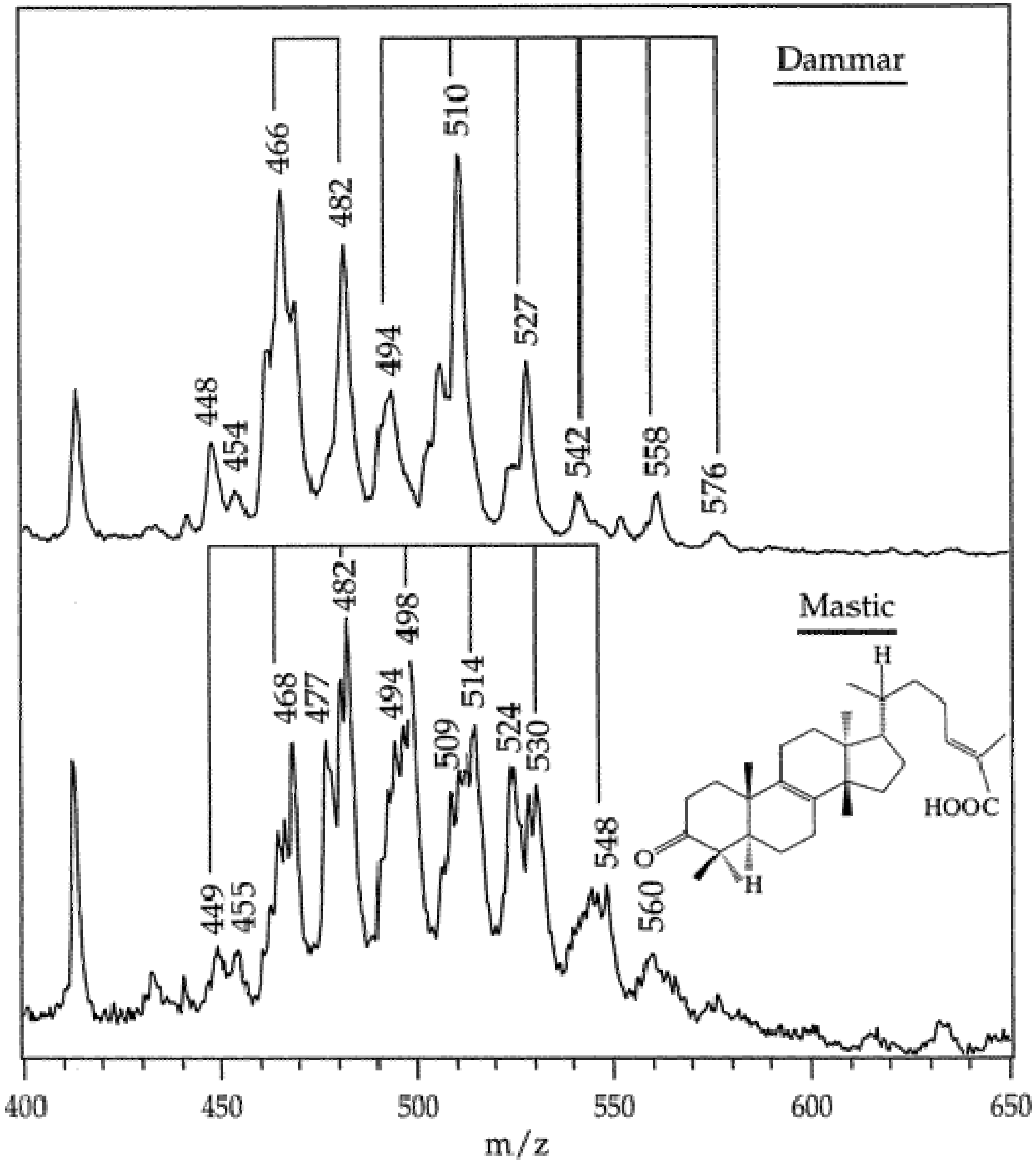
Závěr:

jako pojivo byly v triptychu Benedetta Bonfigliho použity vaječný bílek a žloutek triptych.



MS/MS spektrum *m/z* 402.28, z hydrolyzovaného extraktu z triptychu Benedetta Bonfigliho. Přítomny **y** a **b** fragmenty peptidu vitellogeninu II 50-53 (AGVR).

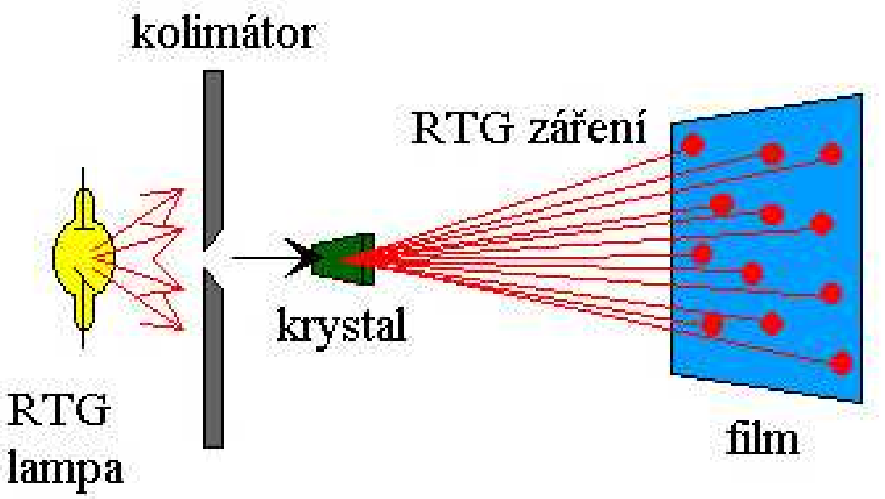
Degradace historických laků



Fotochemická degradace přírodních triterpenoidů použitých jako laky

Hmotnostní spektra laserové desorpce/ionizace dammaru (nahoře) a mastixu (dole) na grafitu.

Rentgenová difrakční analýza (XRD)



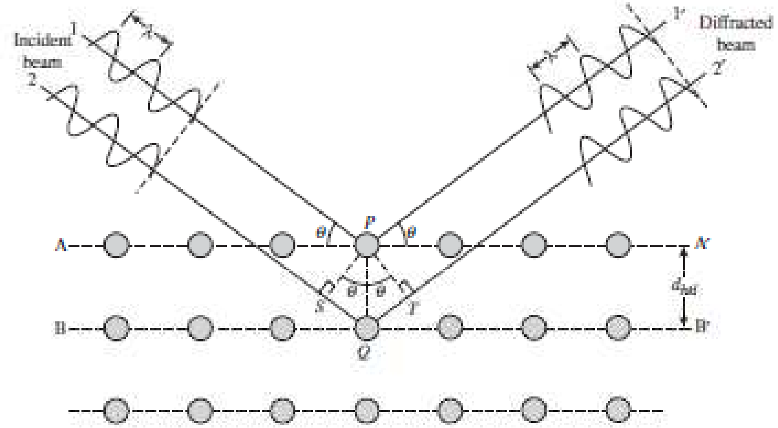


Figure 2.6 Bragg diffraction by crystal planes. The path difference between beams 1 and 2 is $SQ + QT = 2\,PQ \sin\theta$. (Reproduced with permission from W.J. Callister Jr., *Materials Science and Engineering: An Introduction*, 7th ed., John Wiley & Sons Inc., New York. © 2006 John Wiley & Sons Inc.)

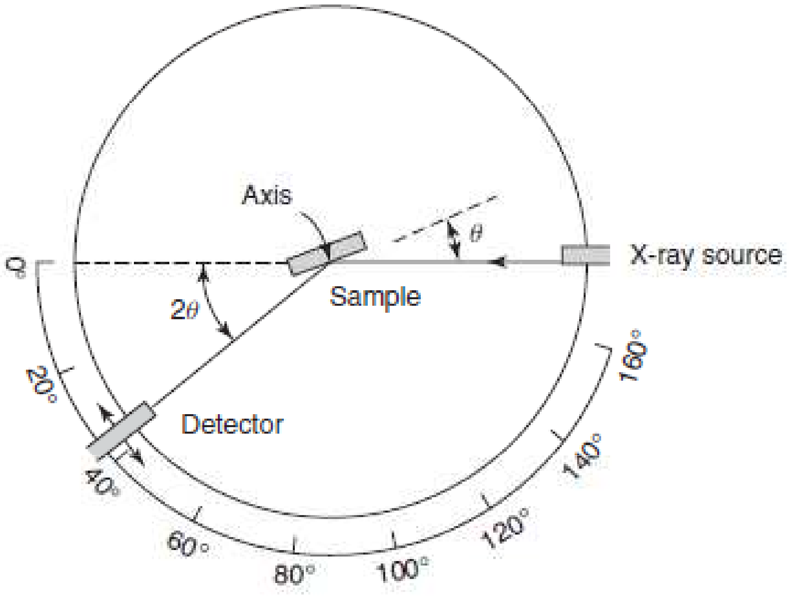


Figure 6.2 Schematic diagram of a XRD diffractometer

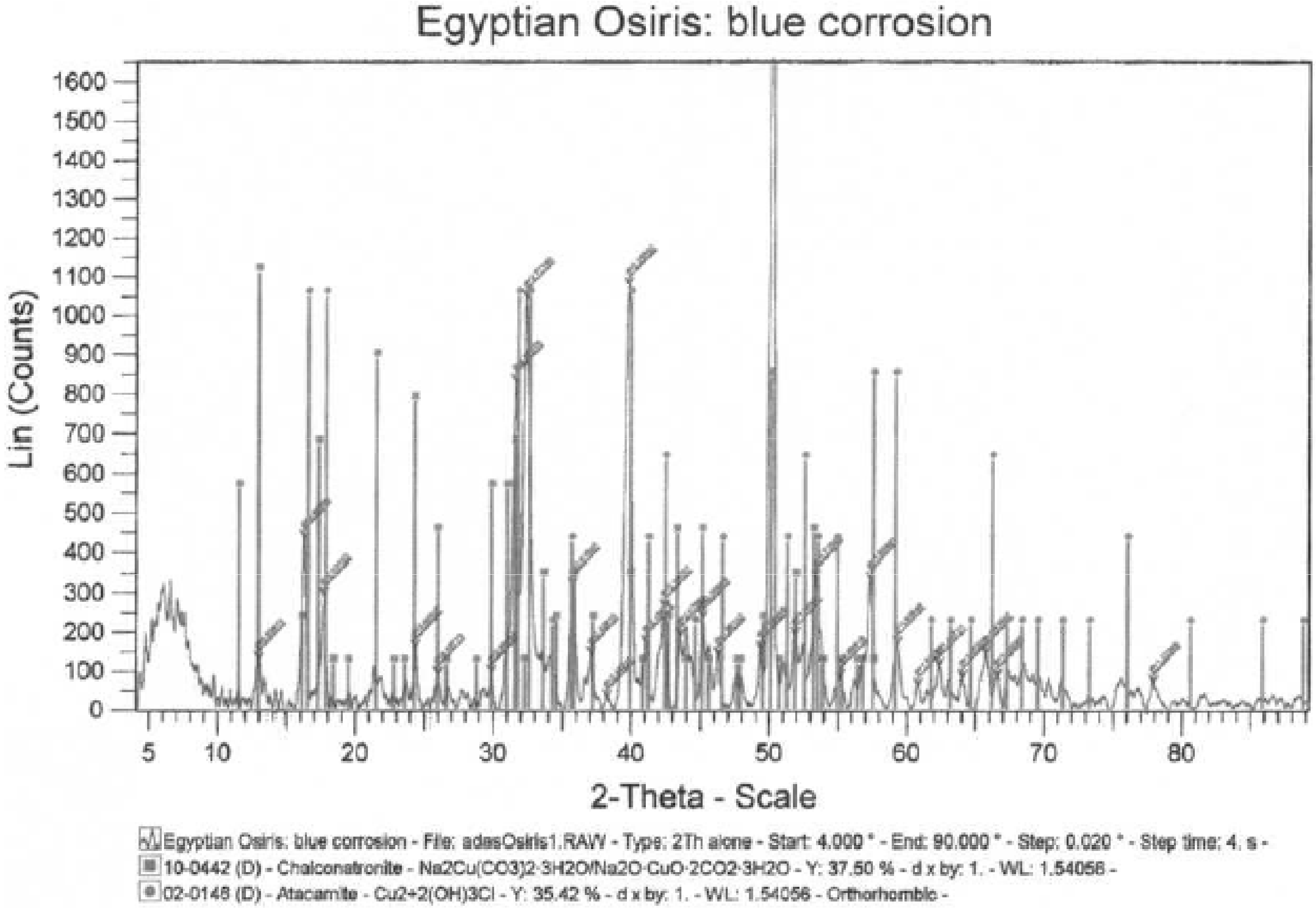


Fig. 10.14. X-ray diffractogram obtained, in situ, for the light blue area of the patina of the Osiris shown in Fig. 10.12, on the back. The principal constituent of this patina is chalconatronite.

Děkuji za pozornost !