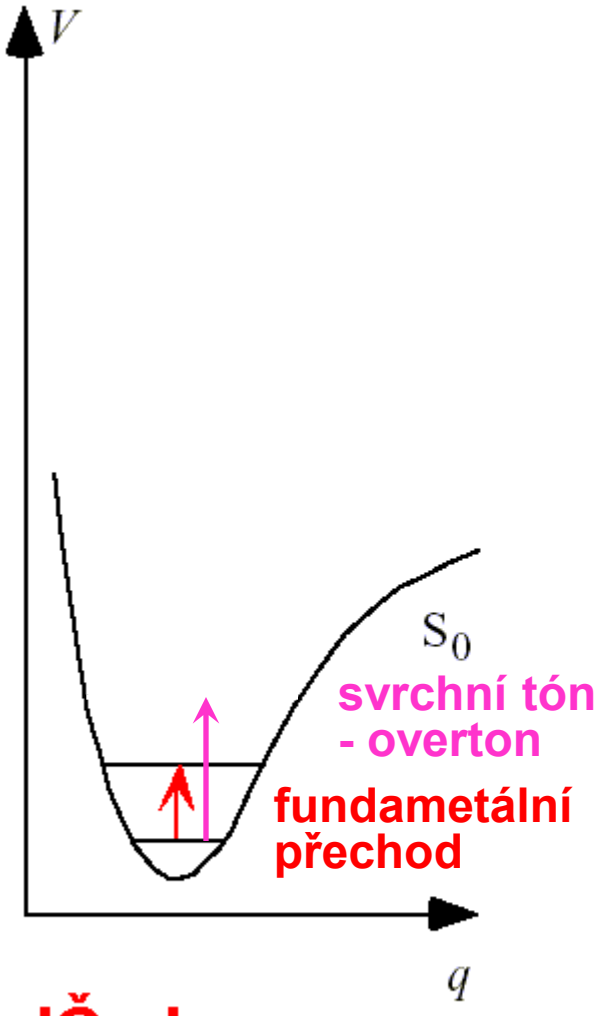
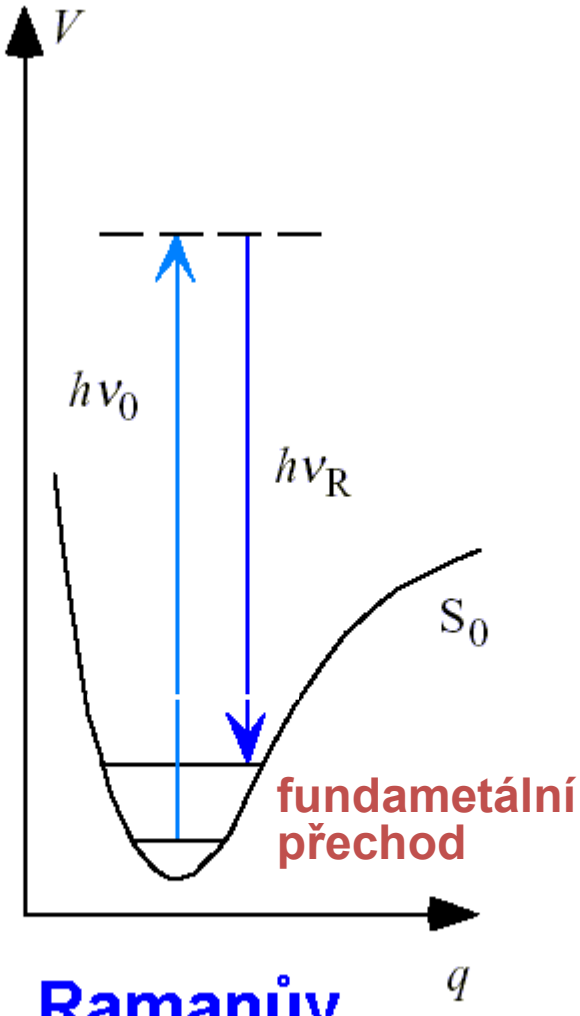
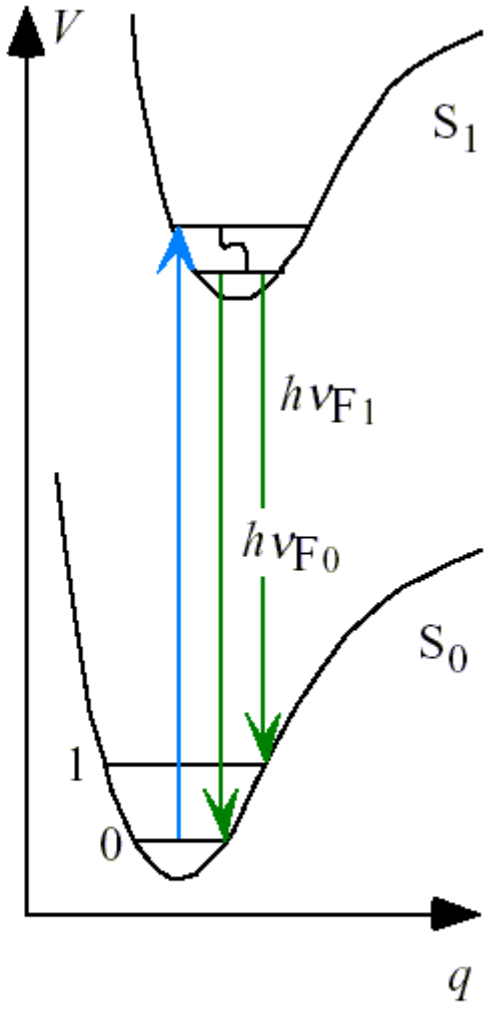
Schéma hladin



IČ absorpce

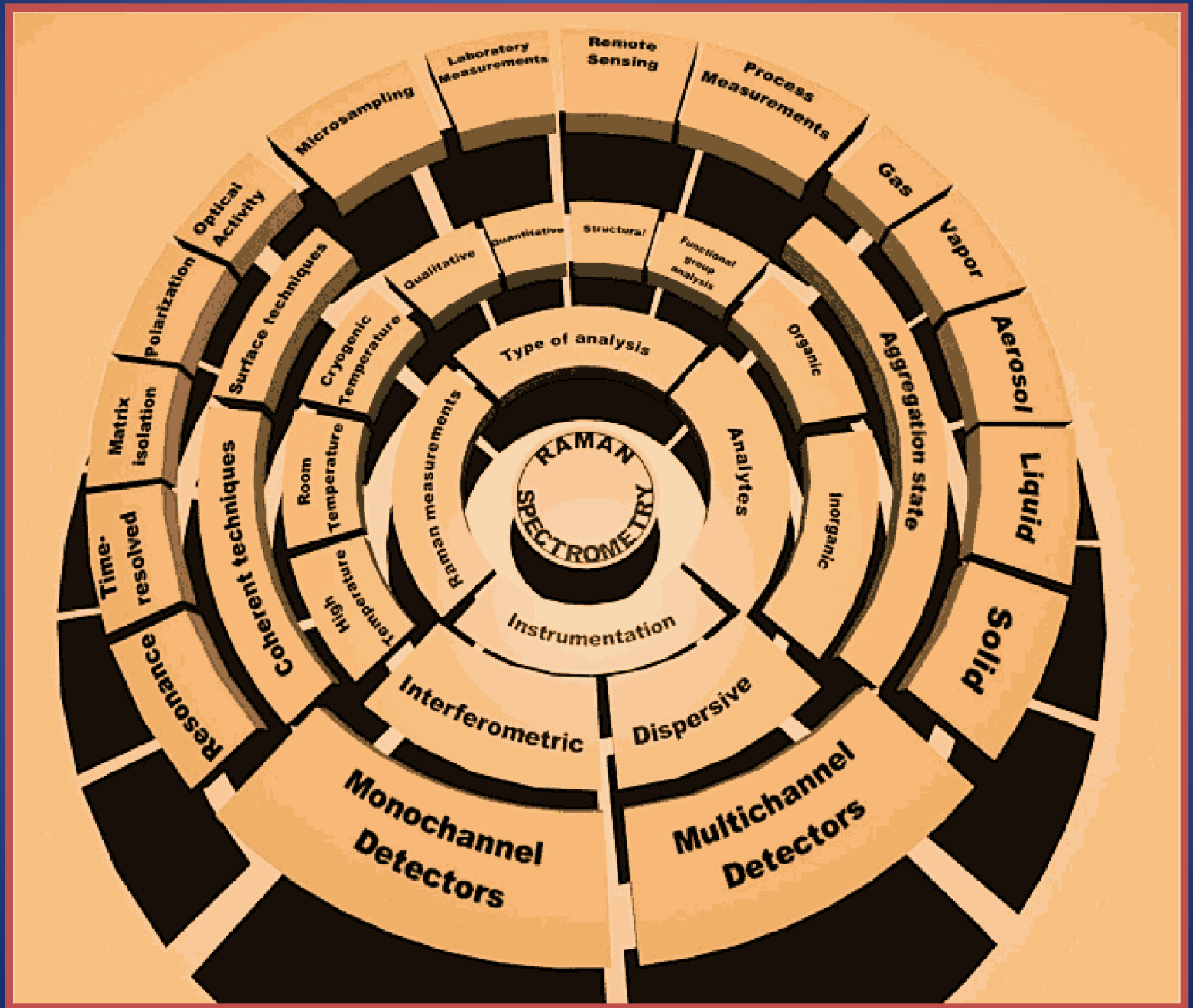


Ramanův rozptyl



Fluorescence

Ramanova spektrometrie



Rozdíly IČ a Ramanovy spektrometrie

Vibrační frekvence jednotlivých módů molekul jsou nezávislé na tom, zda je studujeme infračervenou nebo Ramanovou spektroskopií, avšak intenzity spektrálních linií budou pro obě spektroskopické techniky zřetelně odlišné.

!!!

!!!

Ramanův rozptyl – navíc informace z polarizace/depolarizace rozptýleného záření, z excitačních profilů (rezonanční efekt).

Ramanova spektroskopie

$$\vec{P} = \alpha \vec{E}$$

Klasické přiblížení

- Indukovaný dipólový moment úměrný intenzitě elektrického pole

$$p_x = \alpha_{xx} E_x + \alpha_{xy} E_y + \alpha_{xz} E_z$$
$$p_y = \alpha_{yx} E_x + \alpha_{yy} E_y + \alpha_{yz} E_z$$
$$p_z = \alpha_{zx} E_x + \alpha_{zy} E_y + \alpha_{zz} E_z$$

$$p_k = \alpha_0 E_0 \cos(2\pi \nu_0 t) + \frac{1}{2}\left(\frac{\partial \alpha}{\partial q_k}\right)_0 q_k^0 E_0 \left[\cos(2\pi(\nu_0 - \nu_k)t) + \cos(2\pi(\nu_0 + \nu_k)t)\right]$$

Rayleigh — Stokes — anti-Stokes

Základní výběrové pravidlo Ramanova rozptylu

změna polarizovatelnosti během vibračního pohybu

$$\frac{\partial \alpha}{\partial q} \neq 0$$

Principy Ramanovy a FT Ramanovy spektroskopie

Vztah intenzity pásů

- možnost měření teploty vzorku

$$\frac{I_{\text{anti-Stokes}}}{I_{\text{Stokes}}} = \left(\frac{\nu_0 + \nu_{vib}}{\nu_0 - \nu_{vib}} \right)^4 \mathrm{e}^{-\frac{\mathrm{h}\,\nu_{vib}}{\mathrm{k}\,T}}$$

Instrumentace

The following experiment seems to us to be decisive: between the scattering quartz crystal and the spectrograph slit we placed a quartz vessel which was filled with mercury vapors and totally absorbed light with a wavelength of 2536 A. We did not obtain this line in the spectrogram, but obtained only the satellites.

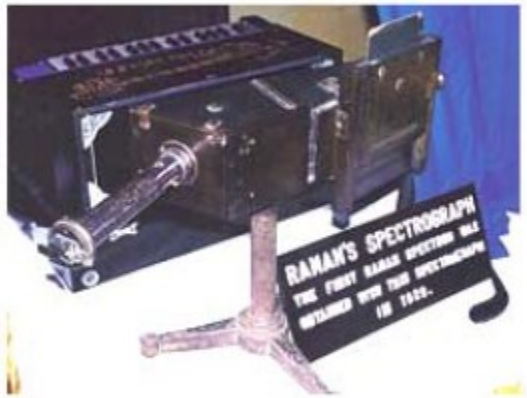
G.S. Landsberg, L.I. Mandelstam, 1928

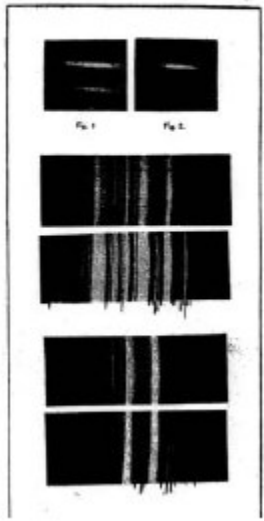
- zdroj excitujícího záření
- excitační optika
- vzorkový prostor
- sběrná optika
- „odlišení“ záření o různé energii
- detekce záření
- akviziční elektronika
- ukládání a zpracování dat

Přehled technik – molekulová analýza

- Techniky Ramanovy spektroskopie
  - Normální RS
  - Resonanční RS
  - SERS, SERRS, TERS
- Ramanova mikrospektroskopie
  - Disperzní
  - FT Ramanova
  - TERS
  - SNOM

Raman's Spectrograph with Photographic Plate and 1st Spectra Published in Indian Journal of Physics

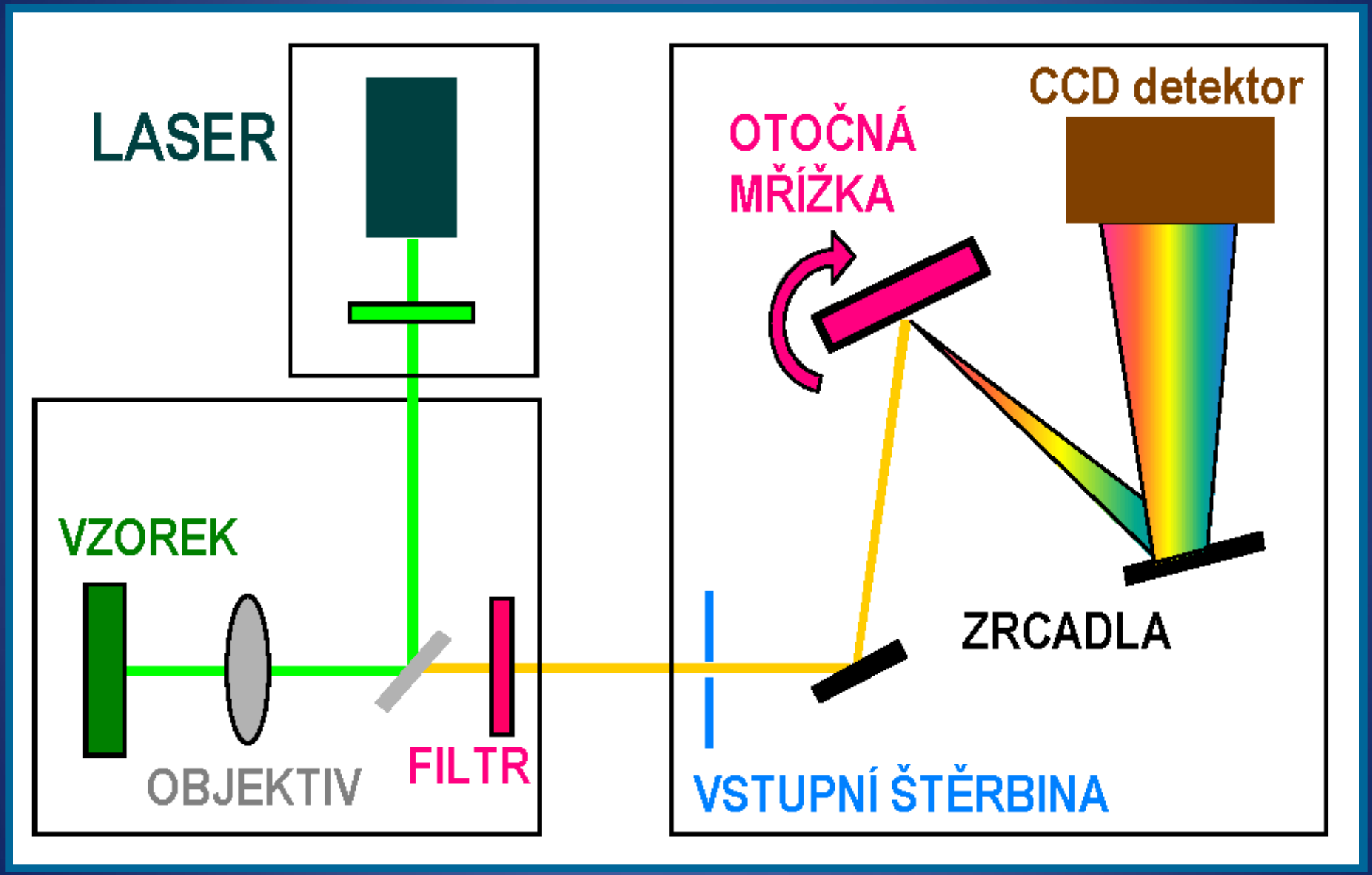




From C&E News, p. 103, January 18, 1999

Externí sondy – vláknová optika

Schéma Ramanova disperzního přístroje



Principy
FT Ramanovy spektroskopie

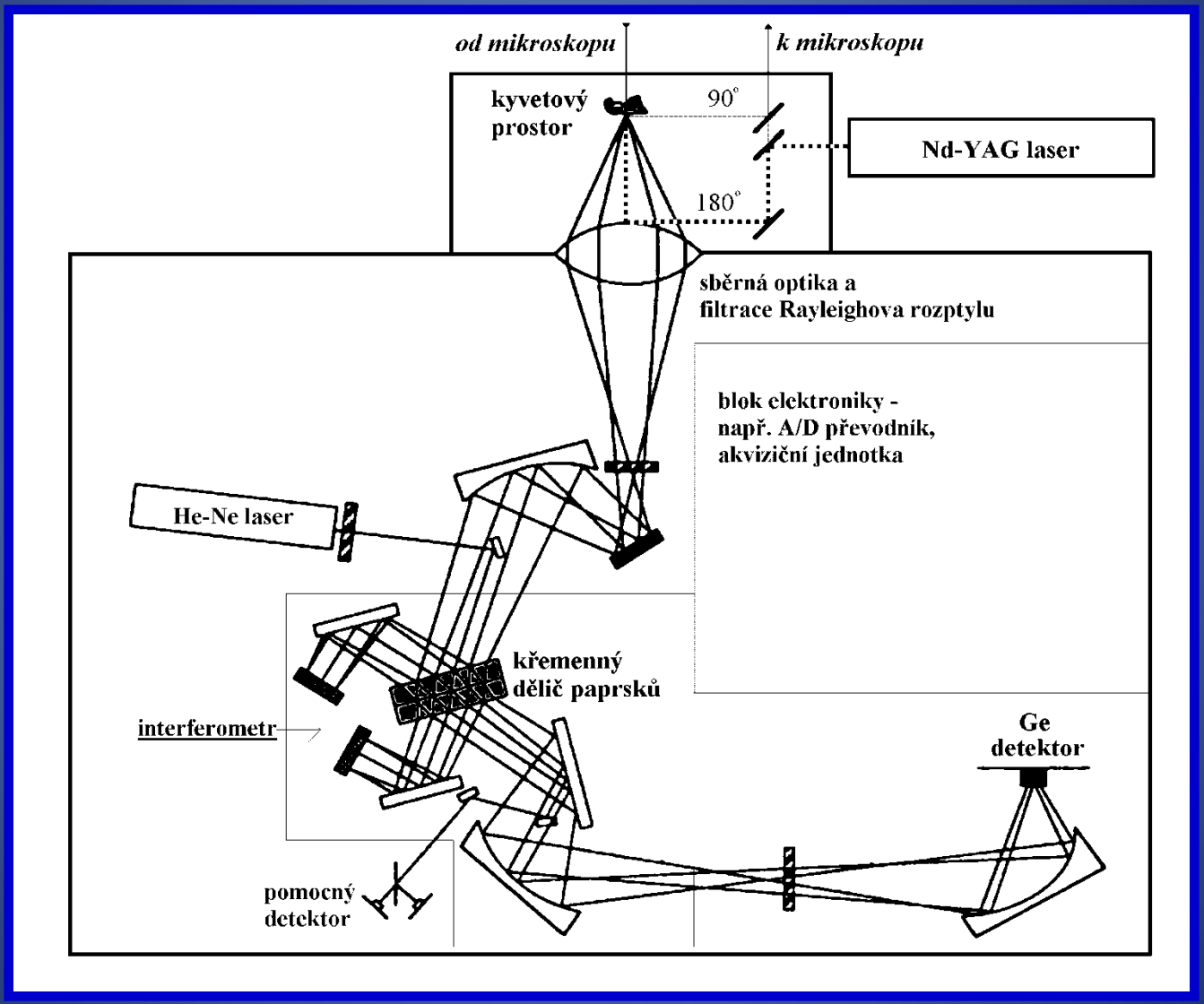


Schéma FT Ramanova spektrometru s NIR excitací

Instrumentace

Materiály pro dělič paprsků

Propustný materiál	Polopropustný povlak	rozsah použití [cm^{-1}]
křemen	Si (Fe_2O_3)	23 000 – 4 000
CaF_2	Si (Fe_2O_3)	10 000 – 1 000
KBr	Ge	4 700 – 350
CsI	Ge	4 000 – 200

Instrumentace

Lasery pro excitaci Ramanova jevu

TYP laseru	vlnová délka [nm]
He-Ne	632,8
Ar^+	514,5
Ar^+	488,0
Ar^+	457,9
Kr^+	568,2
Kr^+	647,1
Kr^+	676,4
Kr^+	752,6
Nd-YAG	1064
Nd-YAG - 2f	532
diodové	780, 785 - NIR
barvivové	360 - 750 - UV, vis

Instrumentace

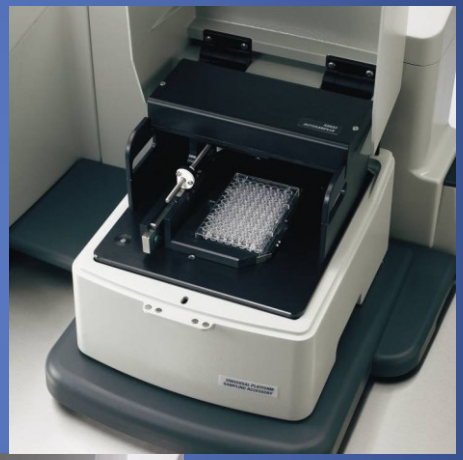
Vzorkování

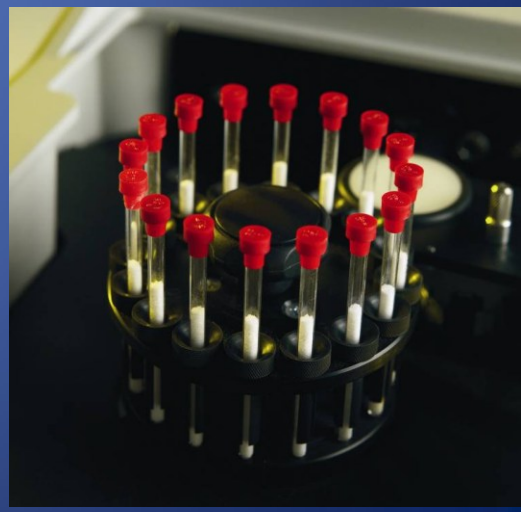
- makroskopické – vialky, skleněné kyvety (NMR, UV-vis …), možnost měřit přes tenkou vrstvu polymeru

Vzorkování

-příklady

-Nicolet





Principy Ramanovy a FT Ramanovy spektroskopie

Problém citlivosti - závislost intenzity rozptylu na vlnové délce

Problém rozlišení - disperzní spektrometry - disperze na mřížce

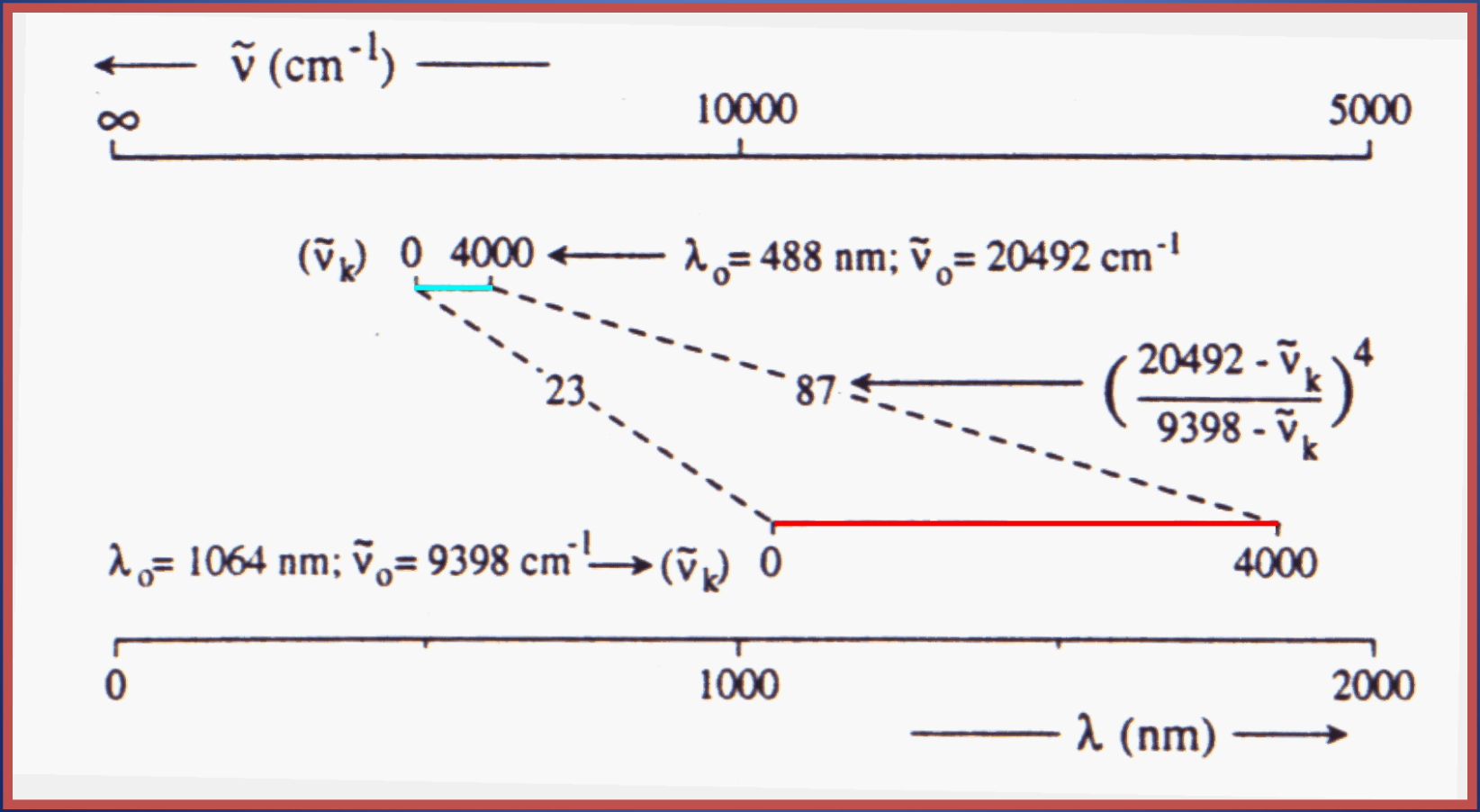
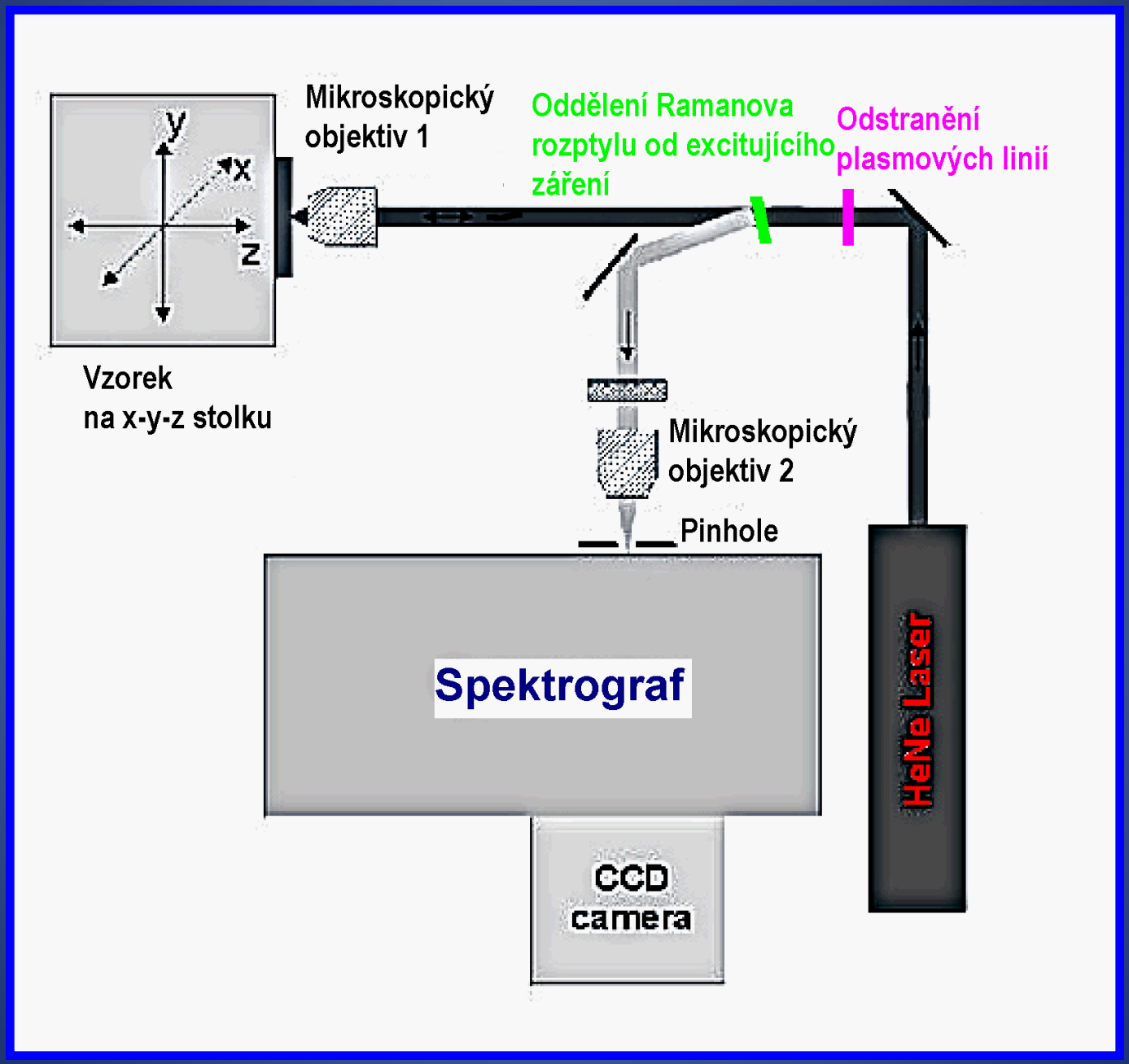


Schéma Ramanova mikroskopu



Ramanův mikroskop

Raman Microscopy Was a microprobe possible?

•In 1966 Delhaye and Migeon argued that a laser beam could be tightly focussed at a sample, and the Raman light efficiently collected and transferred to a spectrometer, without losses.

•Calculations showed that increase in irradiance more than compensated for decrease in size of irradiated volume.

M. Delhaye and M. Migeon, Effet Raman – Calcul du gain apporte par un dispositif optique de concentration du faisceau laser pour l'etude de l'effet Raman, C.R. Acad. Sc. Paris, t.262, 702-705 (7 mars1966)

M. Delhaye and M. Migeon, Effet Raman – Interet de la concentration d'un faisceau laser pour l;excitation de l'effect Raman, C.R. Acad. Sc. Paris, t.262, 1513-1516 (13 juin, 1966)

Tomas Hirschfeld - promoted the idea, based on computations, that sample dimension is not as important as the optical scheme for probing a femto liter sample. His ideas appeared as an abstract Hirschfeld J Opt. Soc. V63 1973

Ramanův mikroskop

Evolution of Microspectroscopy

1974 **Fourth International Conference on Raman Spectroscopy, Brunswick, ME, - reports of first prototypes of 1973 based on doubles with PMT's**

#5.1.10 GJ Rosasco and E Etz, Investigation of the Raman Spectra of Individual Micron Sized Particles

#5.B M Delhaye and P Dhamelincourt, Laser Raman Microprobe and Microscope

1980's **Triple spectrographs with microscopes and IPDA's**

1989 **Raman microscopes with CCD multichannel detector**

1991 **Notch filters used in injection/rejection of laser**

Rebirth of widefield, direct (global) imaging due to use of optical tunable filters (AOTF, LCTF, Interference)

Introduction of confocal microscopy for point analysis and imaging (point and linescan) providing improved

- **depth resolution**
- **contrast of Raman images/maps**
- **rejection of fluorescence**

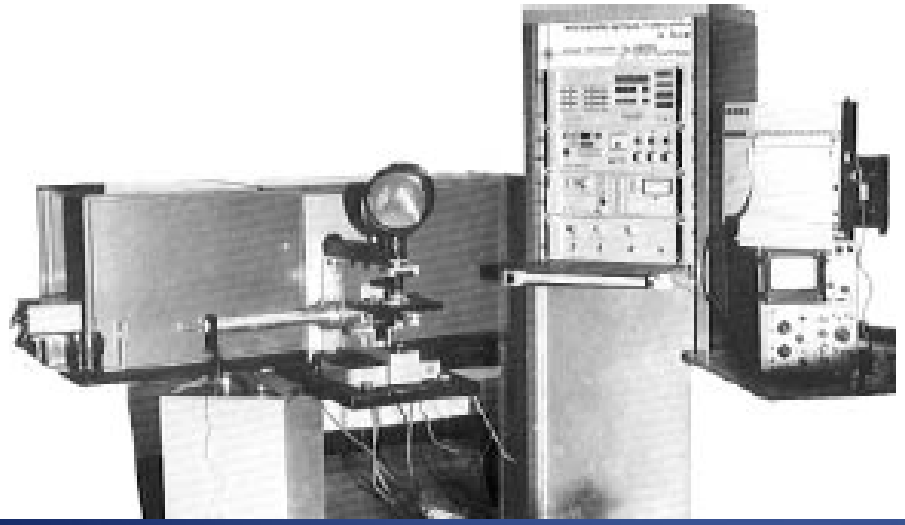
1992 **European patent for confocal line-scanning**

Ramanův mikroskop

MOLE™ (Molecular Optics Laser Examiner)

First Commercial Instrument - ca. 1974

- Double spectrometer/spectrograph
- Imaging implemented with global illumination and sample re-imaged onto gratings and then detector
- Many early applications were in Geosciences and contaminant analysis in industrial materials.
- Lack of sensitivity made Raman mapping/imaging impractical due to high background levels; problems eventually overcome with confocal coupling and data treatments including multivariate algorithms.

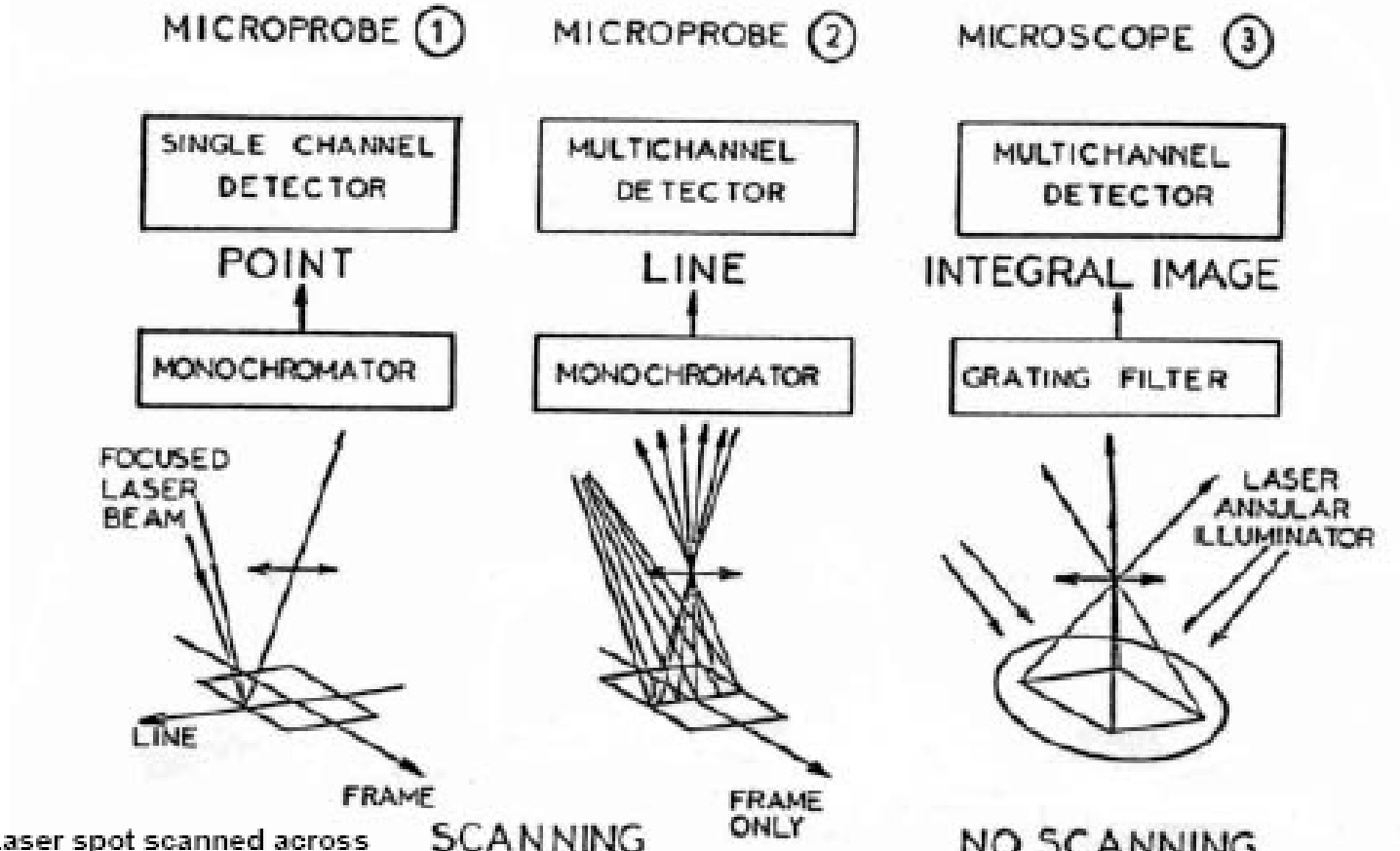


Prototype - L'Universite des Sciences et Techniques de Lille France

Ramanův mikroskop

Original Concepts of Raman Microscope

Initial goal was to produce a picture of the sample through its Raman signal in order to promote the development to the Raman community.



Laser spot scanned across sample - image reconstructued from PMTsignal

Laser spot scanned along line and imaged thru stigmatic mono onto multichannel detector

Global laser illumination - sample imaged onto detector thru λ-selection filter

Delhaye and Dhamelincourt, J Raman Spectros. 3, 33-43 (1975)

Ramanův mikroskop

Solid Inclusion in Mineral

White light Illumination | Sulfur line at 473 cm^{-1} | $SrSO_4$ line at 1000 cm^{-1}

These images were recorded on the MOLE™ using Global Imaging and published in product literature in 1976.

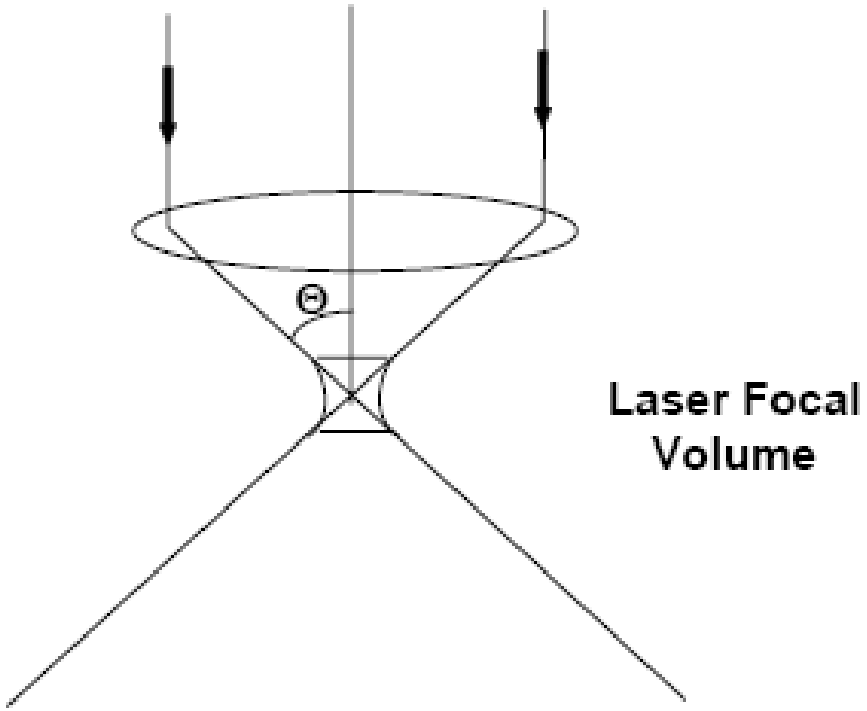
Ramanova mikrospektroskopie

Laser Focus by Microscope Objective

The availability of high na objectives enable the small focus

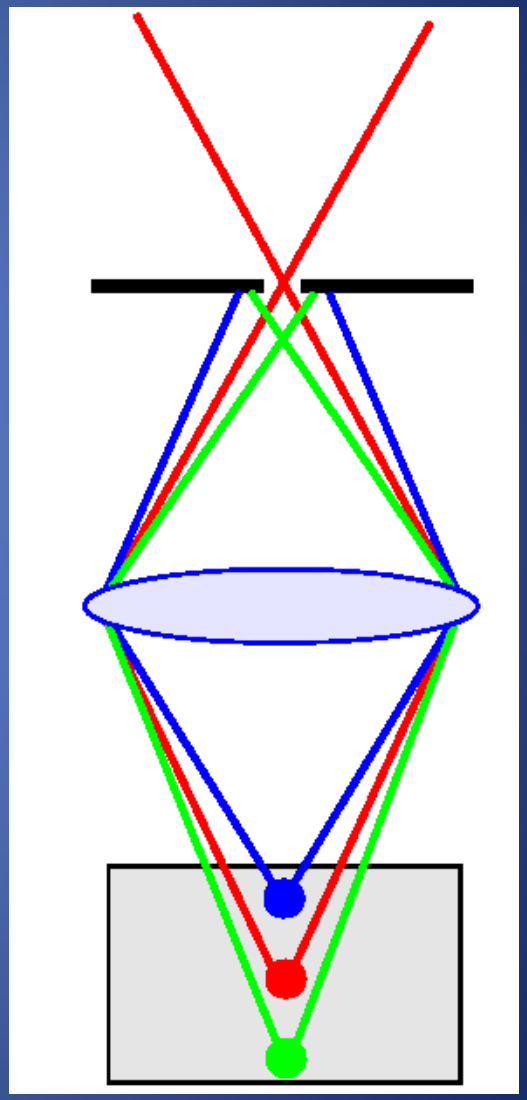
$$\omega_o \cong \lambda/na \Rightarrow 0.5 - 1\mu m$$

$$na = n \sin\Theta$$



Ramanova mikrospektroskopie

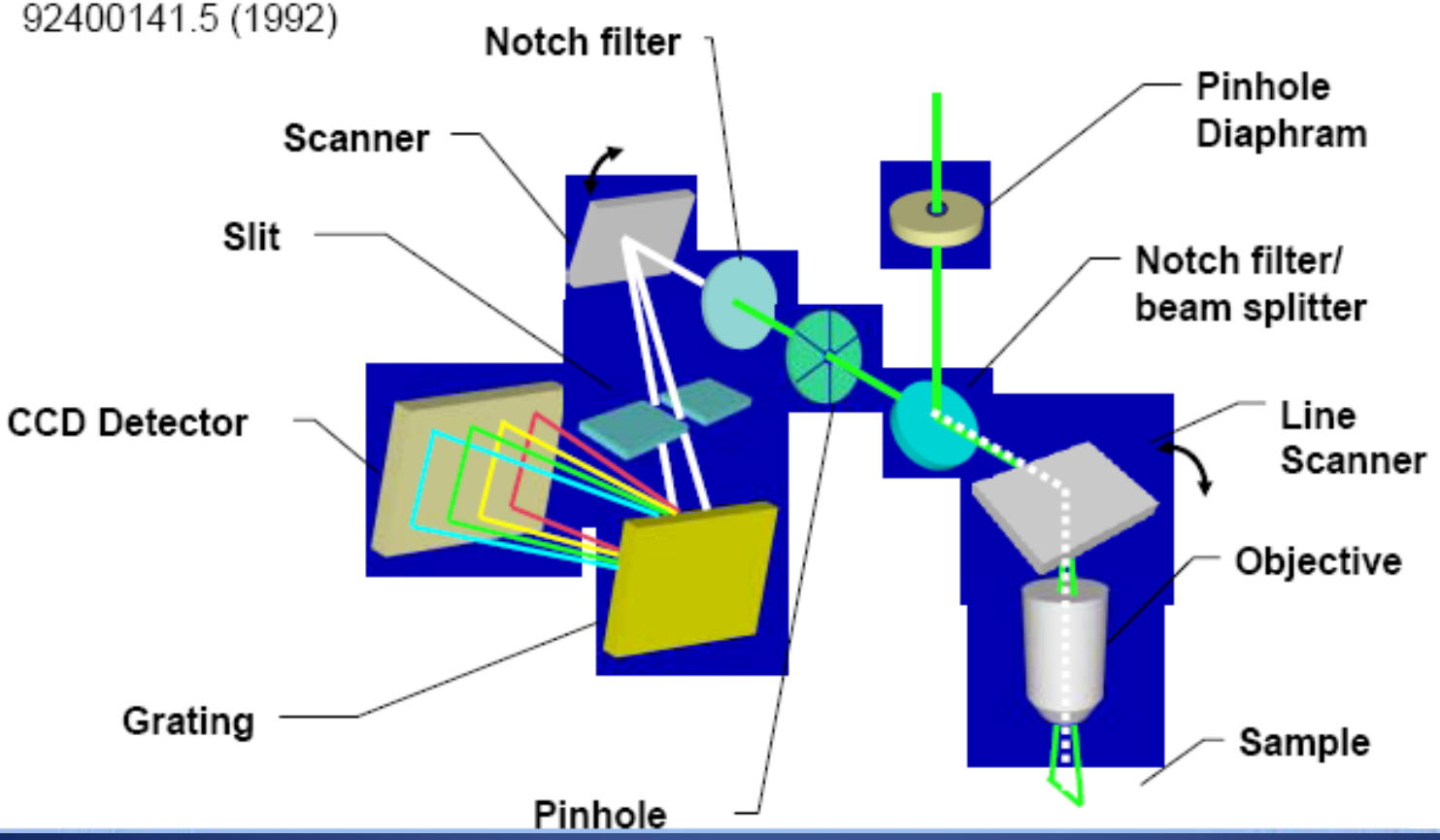
- Dispersní
  - viditelná excitace
  - možnost konfokálního režimu pro lepší prostorové rozlišení
- FT Ramanova
  - NIR excitace
  - horší prostorové rozlišení
  - menší riziko fotorozkladu a fluorescence



Ramanova mikrospektroskopie

Confocal Line Scanning

Patented unique method to scan laser through objective to avoid aberrations and maintain diffraction-limited spot. Then the spectrum of each spot on sample is displayed on a different row of the CCD, **assuming that the spectrograph is stigmatic.** European patent # 92400141.5 (1992)



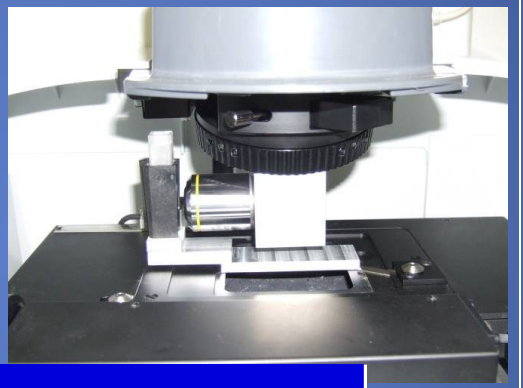
Instrumentace

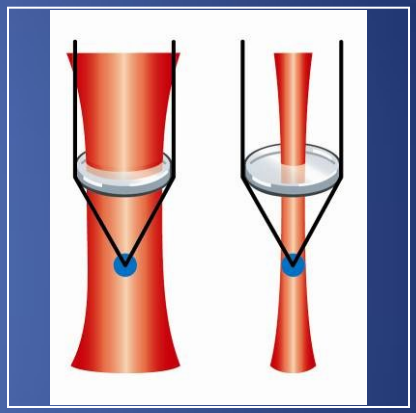
Vzorkování

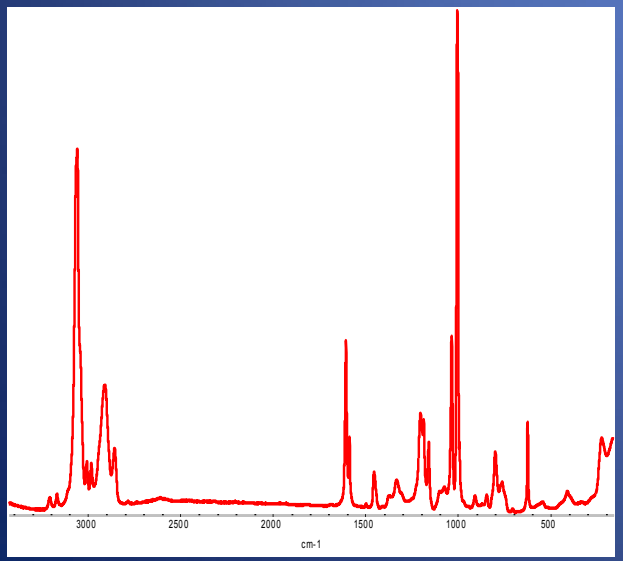
- mikroskopické – objektivy s různým zvětšením, nástavce pro makro měření

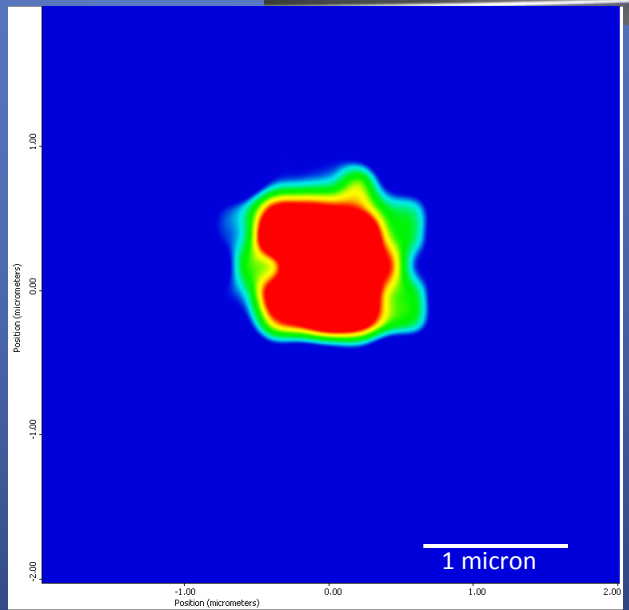
Vzorkování

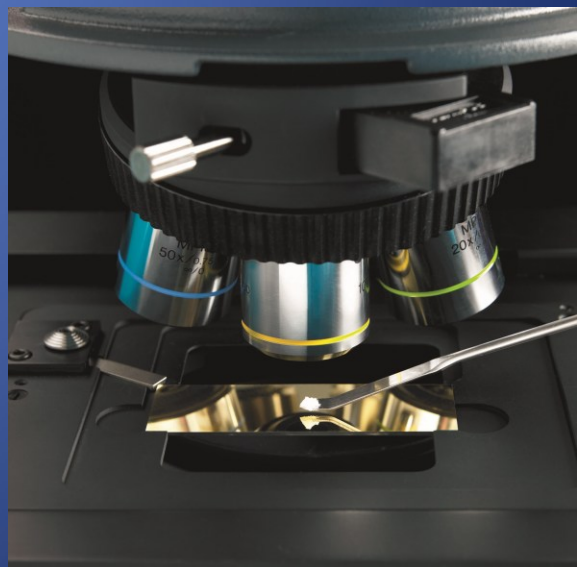
- mapování povrchů
- volba sledované plochy
- konfokalita – hloubkový profil
- autofokus











Raman Microspectrometers

Renishaw

Kaiser Optical

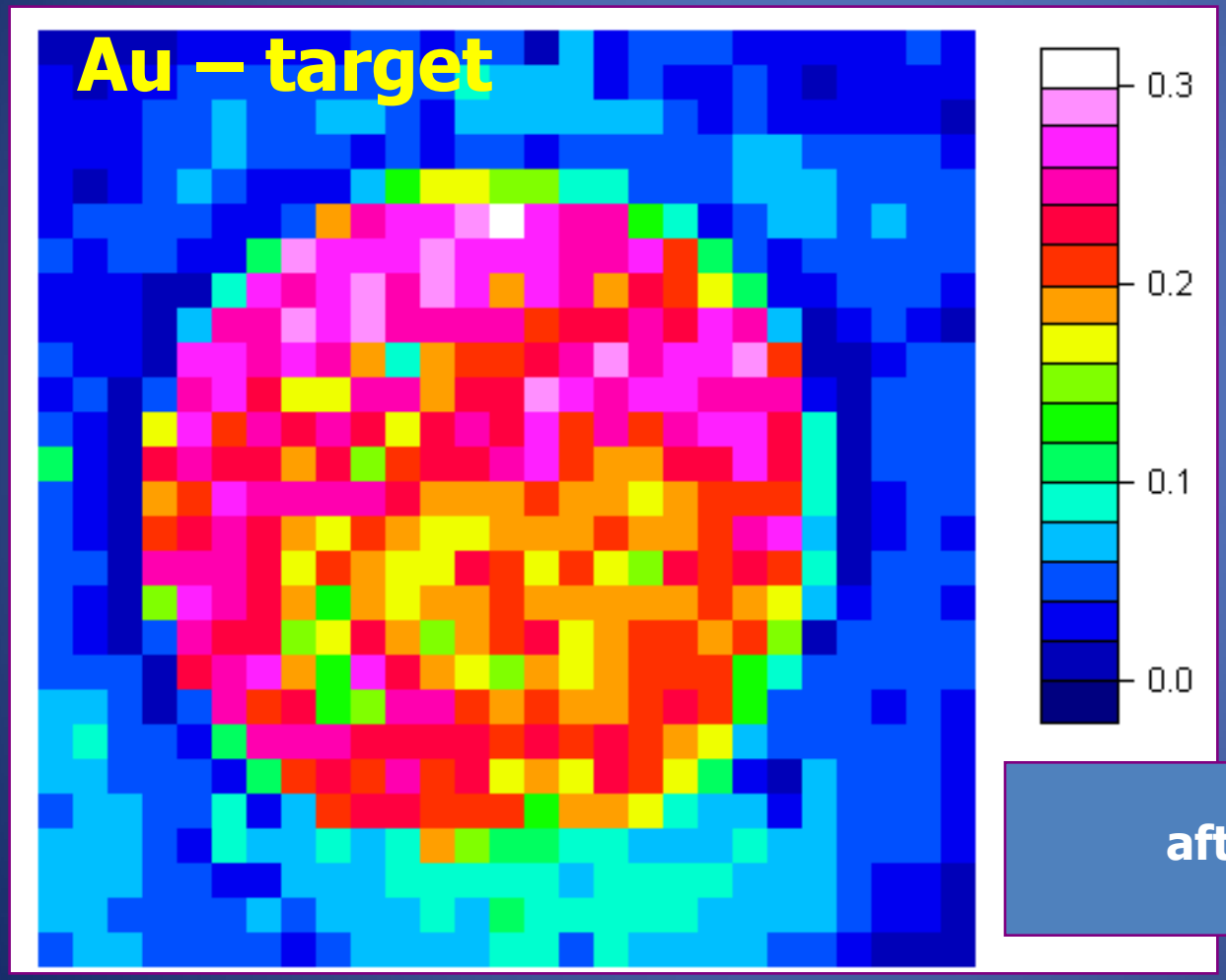
Chromex

Nicolet

Jobin-Yvon

Raman Microspectroscopy

- mapping of surfaces - Gram-Schmidt

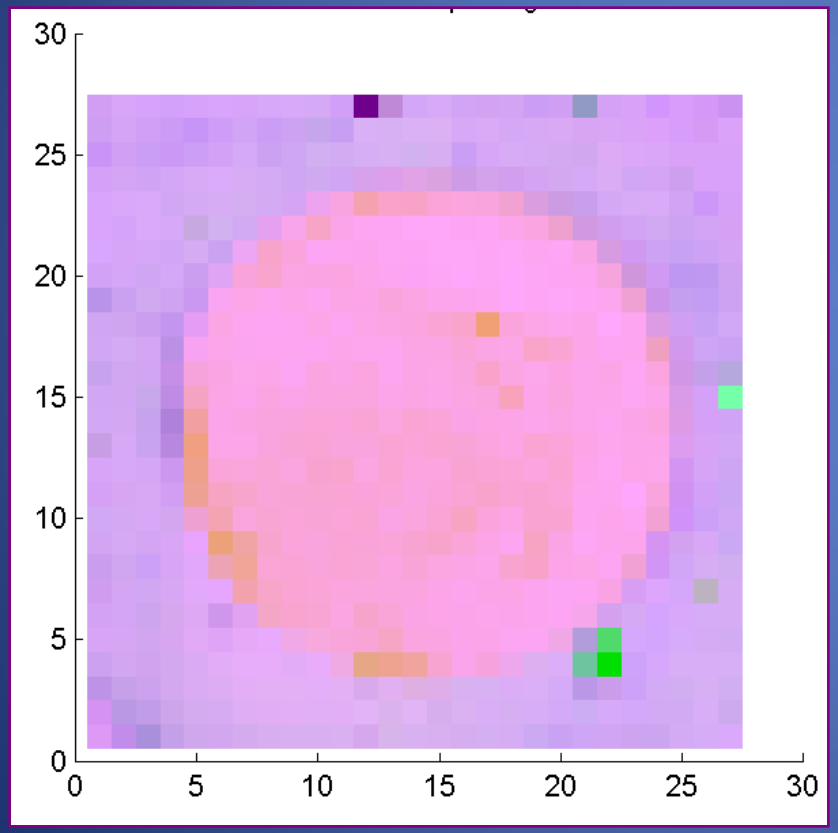


after deposition of 4-aminothiophenole

Raman Microspectroscopy

- mapping of surfaces – PCA (RGB)

Au – target

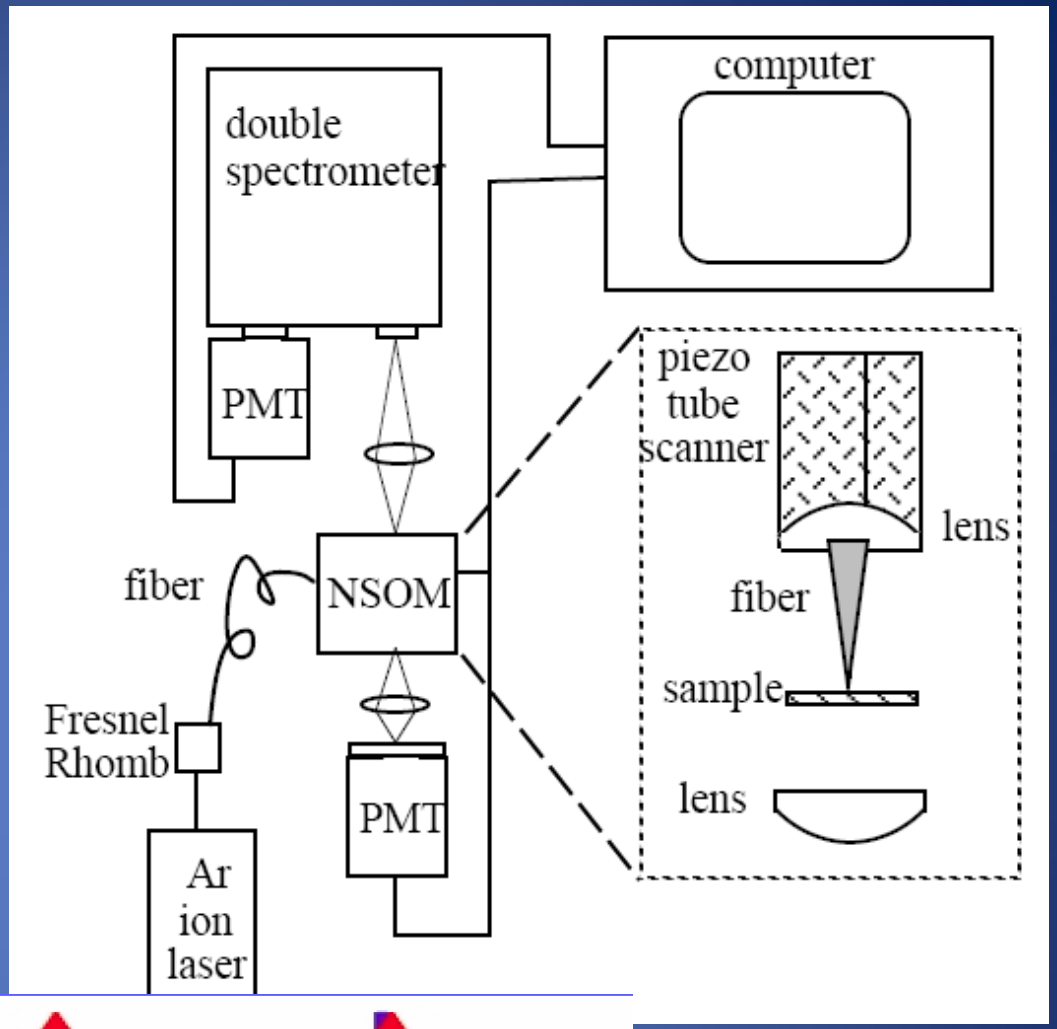


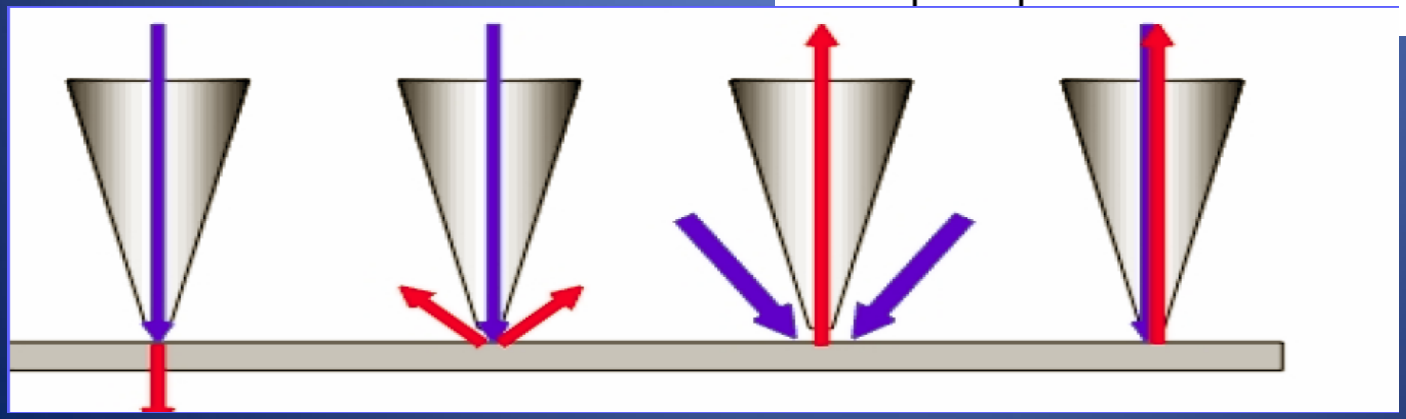
red – value of 1-st principal component (PC)
green – value of 2-nd PC
blue – value of 3-rd PC

after deposition of 4-aminothiophenole

Raman - NSOM

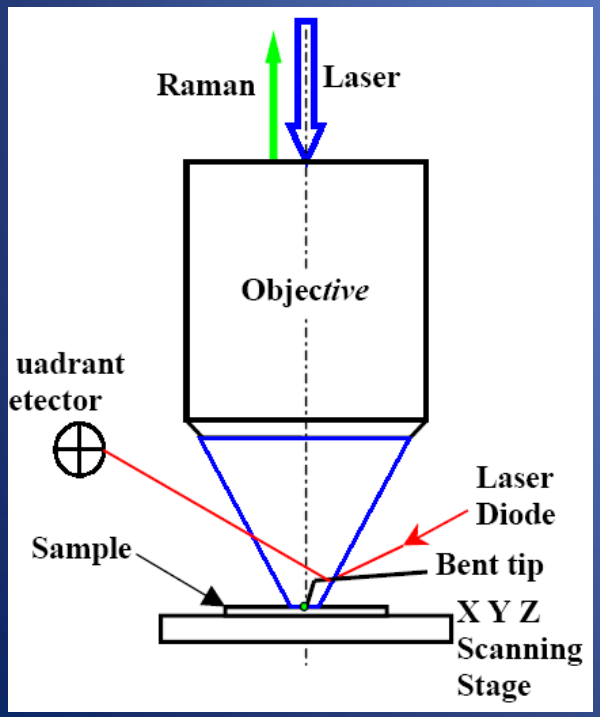
- vzdálenost sondy – do 10 nm
- apertura sondy
- režimy snímání
  - transmisní (jen transparentní vzorky)
  - reflexní – ostrá sonda – vysílač, přijímač, obojí
  - rozptyl – vysílač, přijímač, obojí

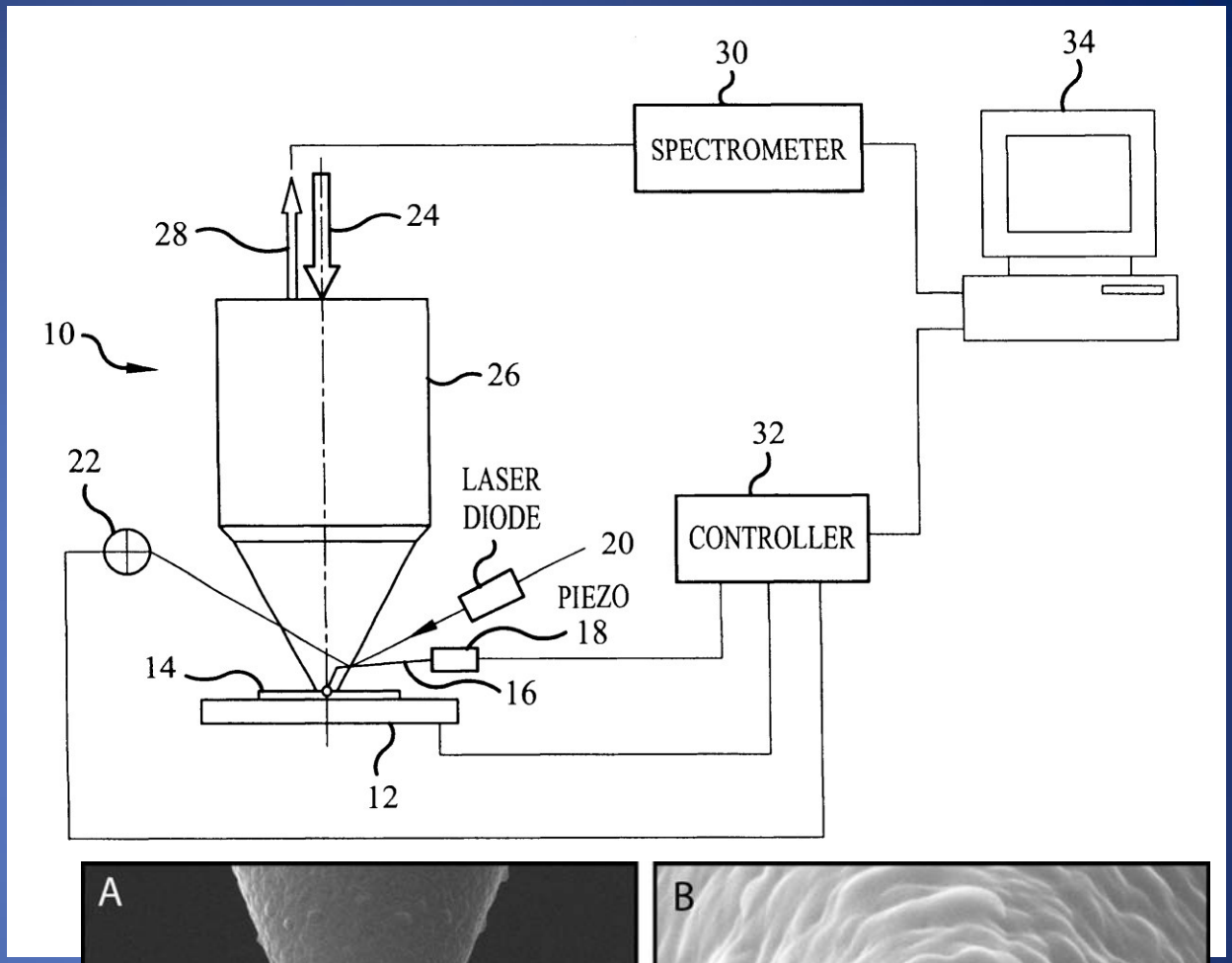


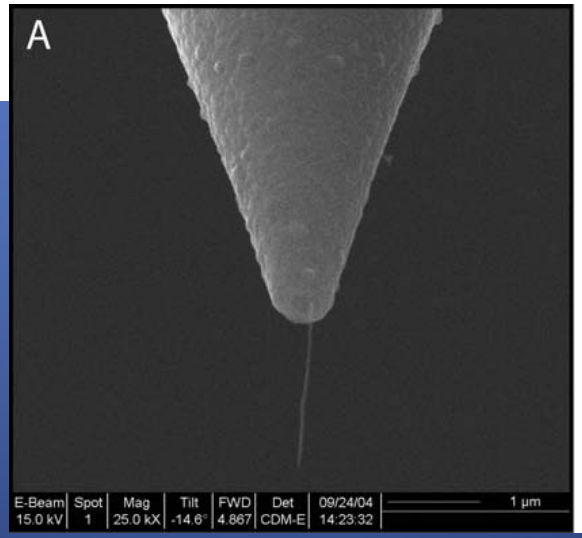


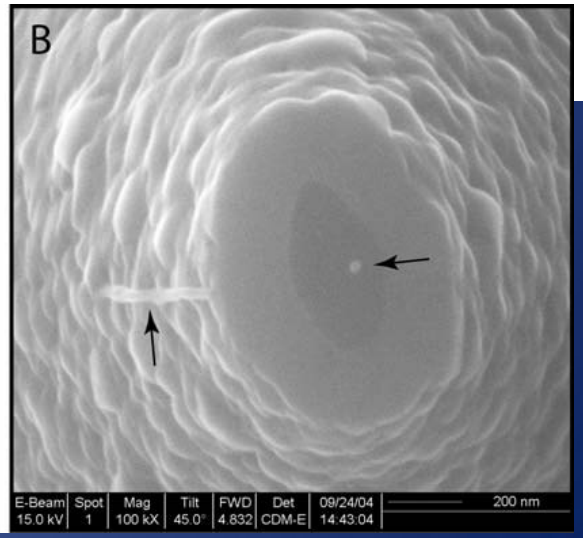
Raman - NSOM

- vzdálenost sondy – do 10 nm
- apertura sondy
- režimy snímání
  - kolmá či šikmá laserová excitace







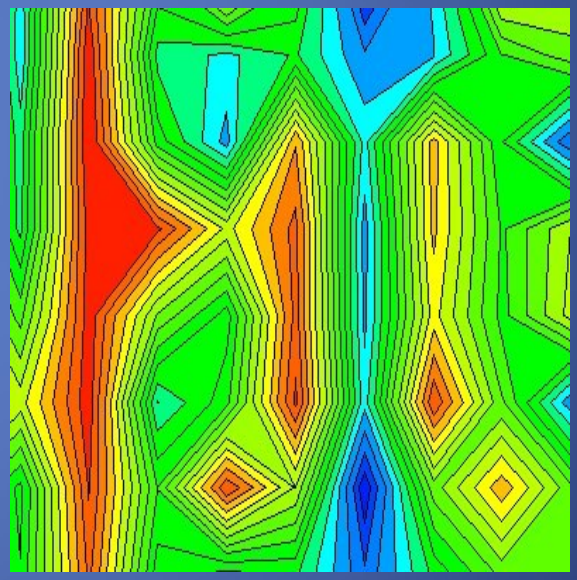


Combination

– MicroRaman, SNOM enhanced Raman, AFM

Parallel imaging of a Silicone Semiconductor

AFM image – 9 x 7 μm



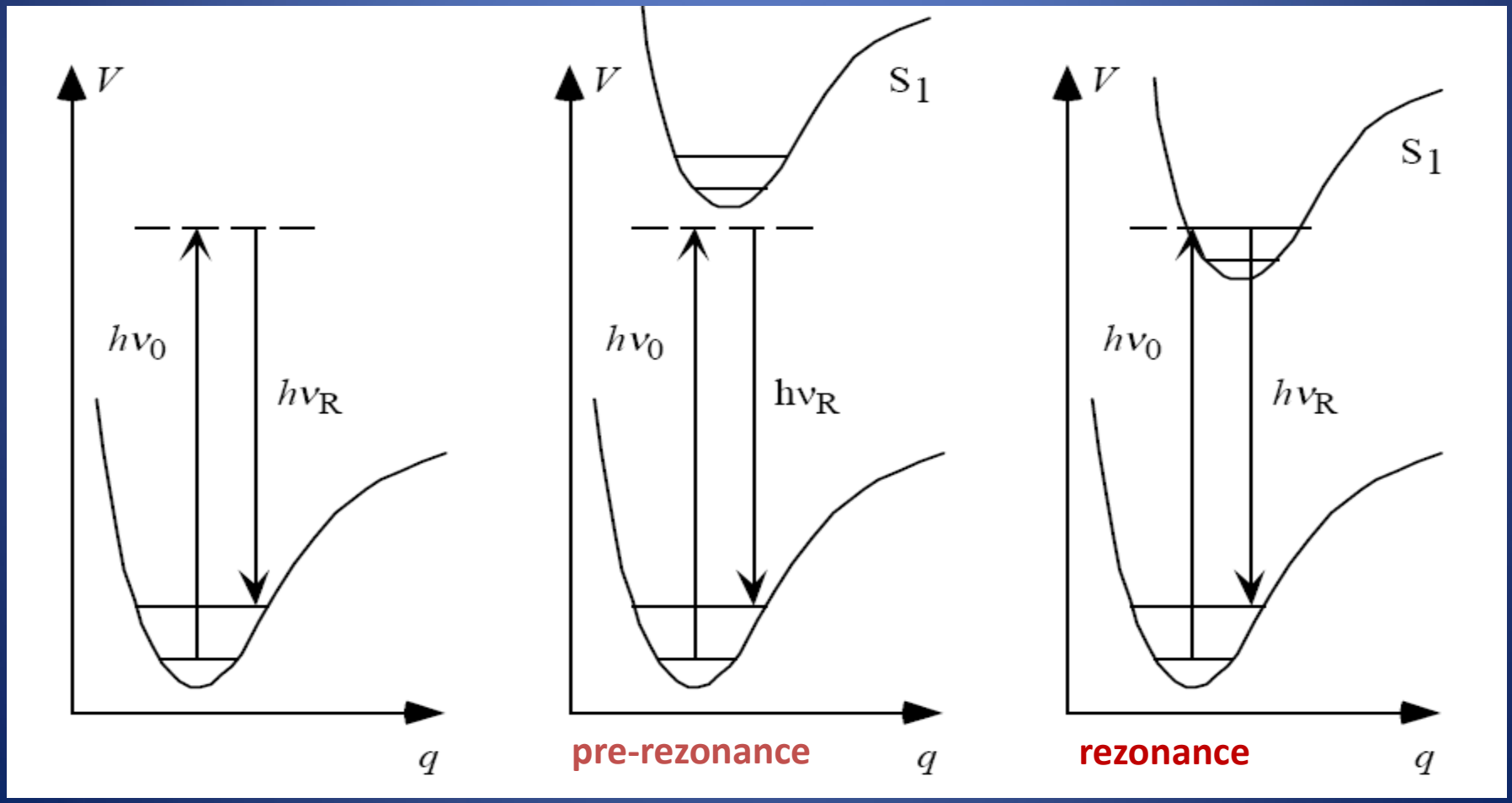
Raman instensity image – 520 cm^{-1}, *the same area*

Speciální techniky

- rezonanční - **RR**
- povrchem zesílený - **SERS**
- rezonanční povrchem zesílený – **SERRS**
- hrotem zesílený – **TERS**
- *fotoakustický* - **PARS**
- *časově rozlišený* – **TRRS**
- *Ramanova optická aktivita* - **ROA**
- **hyperRaman**
- koherentní anti-Stokes - **CARS**
- koherentní Stokes - **CSRS**

Rezonanční Ramanův rozptyl

- virtuální hladina a elektronově excitované stavy



Rezonanční Ramanův rozptyl

- virtuální hladina v blízkosti elektronově excitovaného stavu
  - UV rezonanční Ramanova spektroskopie – nukleové kyseliny, proteiny
  - Viditelná oblast – koordinanční sloučeniny, organická barviva, hemoproteiny
  - NIR – „prerezonance“ ? – nízkoenergetické elektronové přechody

  - Excitační profily – závislost Ramanových spekter (vybraných pásů) na excitační vlnové délce

Rezonanční Ramanův rozptyl

- Faktor zesílení – 10^2 – 10^4
- Franck-Condonovo zesílení
  - Složka normální souřadnice vibrace je ve směru „expanze“ molekuly během elektronové excitace (dýchací vibrace porfyrinového makrocyklu)
- Vibronické zesílení
  - spřažení se dvěma excitovanými stavy

Rezonanční Ramanův rozptyl

- Faktor zesílení – 10^2 – 10^4
  - Praktické aspekty

  - U roztoků – otázka volby koncentrace a pozice excitačního paprsku
    - Samoabsorpce
    - Fluorescence

    - Volba geometrie paprsku, fokusace
    - Koncentrační profil

Rezonanční Ramanův rozptyl

- Závislost rezonance na velikosti uhlíkových nanotrubic

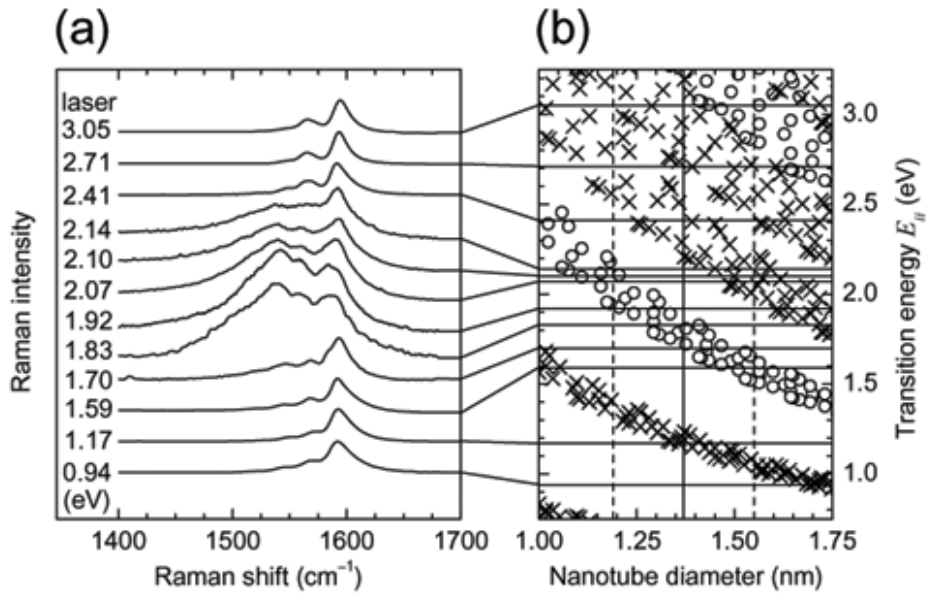


Fig.1– (a) Raman spectra of the tangential G-band modes of SWNT bundles measured with several different laser lines, on a sample with $d_t = 1.37 \pm 0.18$ nm (Pimenta et al. 1998). (b) Resonant transition energies E_{ii} *vs.* d_t. The vertical solid line is the average d_t and the vertical dashed lines denote the d_t distribution width. Crosses are for S SWNTs and open circles for M SWNTs (Samsonidze et al. 2003).

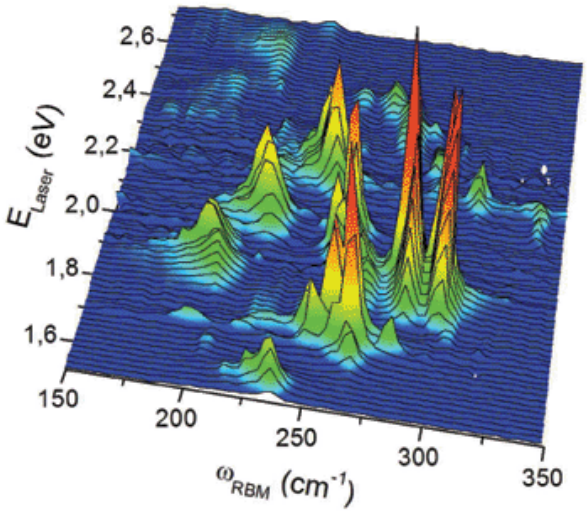


Fig. 4 – RBM Raman measurements of HiPco SWNTs dispersed in SDS aqueous solution, measured with 76 different laser lines (Fantini et al. 2004). The nonresonance Raman spectrum from a separated CCl_4 solution is acquired after each RBM measurement, and is used to calibrate the spectral intensities and to check the frequency calibration. Each Raman peak comes from the radial breathing mode (RBM) of a specific (n, m) single-wall carbon nanotube that enters and leaves resonance. The RBM frequencies are inversely proportional to the nanotube diameters.

Studované materiály

VZORKY – pevné látky, kapaliny, fázové rozhraní

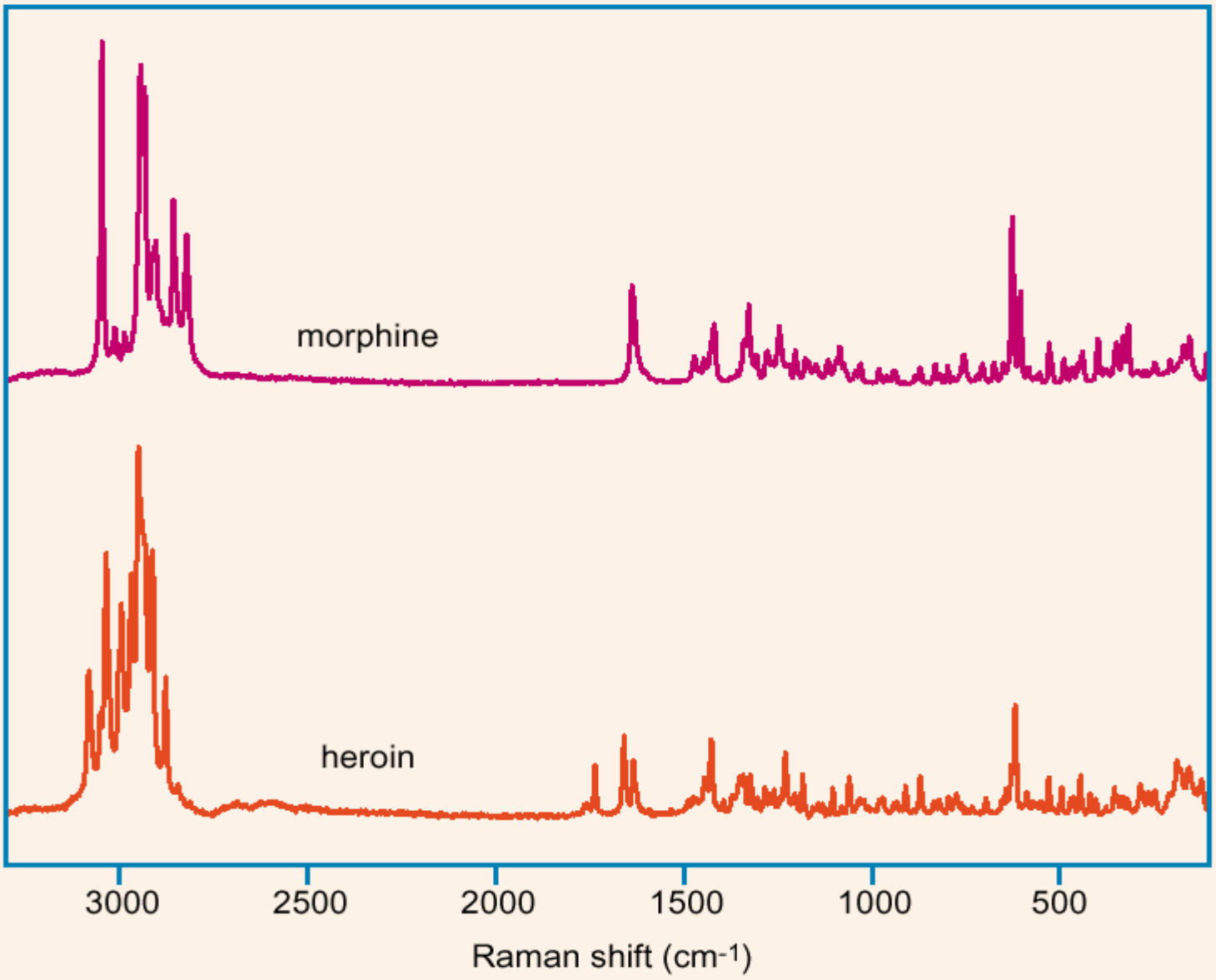
- příklady

- **anorganické -** *korozní vrstvy,* povrchy pevných disků, křemík, *amorfní uhlík, diamanty*
- **organické -** *supramolekulární systémy, kontaminanty v životním prostředí*
- **polymery -** *fotolabilní materiály*
- **biologické** - *in vitro, in vivo*
- **geologické -** *minerály, horniny*
- **archeologické -** od paleolitu po novověk

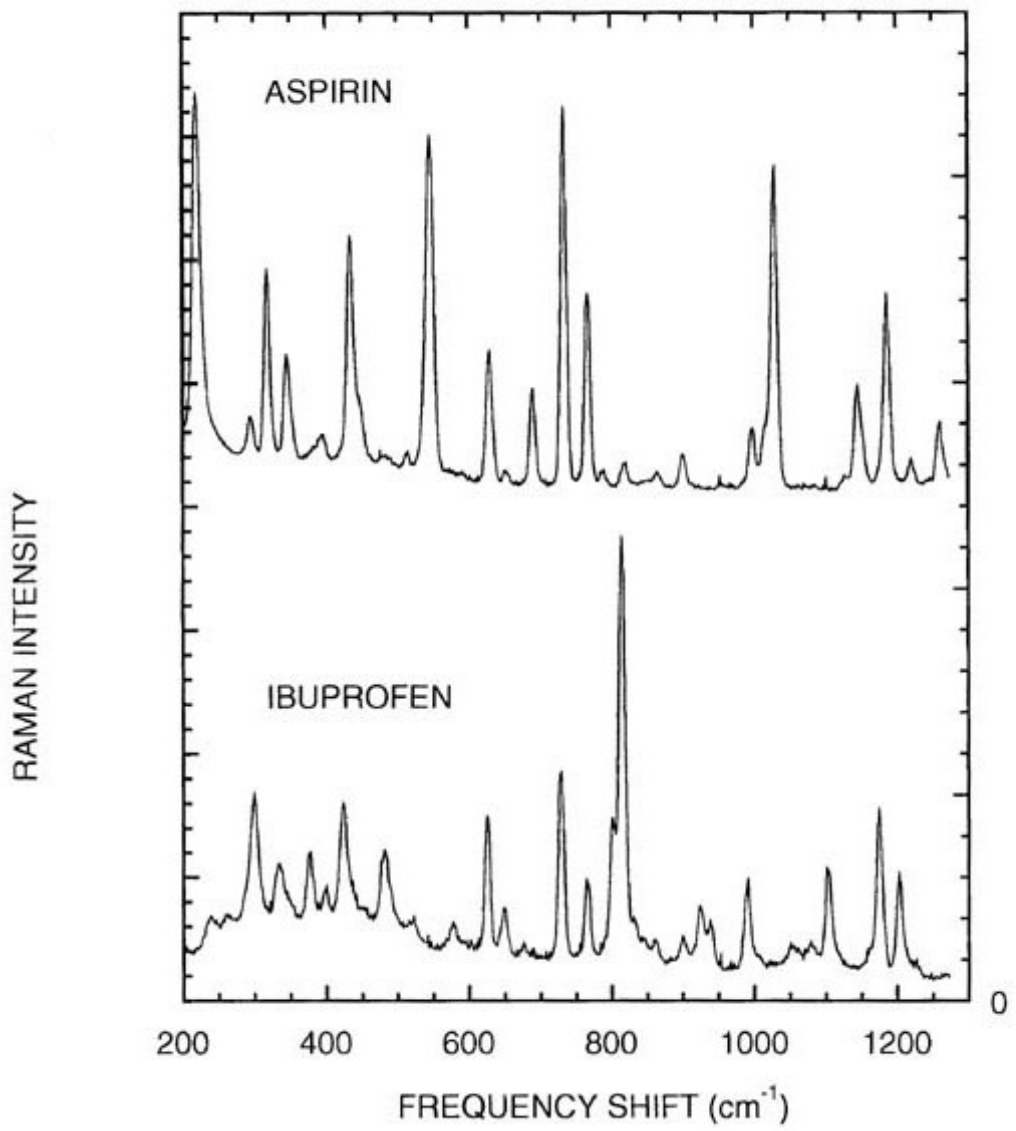
Použití spektrálních dat

- Interpretace spekter – strukturní analýza, identifikace látek – spektrální knihovny
- Intenzita pásů – kvantitativní analýza
- Časově rozlišená spektra – kinetické studie
- Teplotně závislá spektra
- Analýza směsí – identifikace subspekter – faktorová analýza

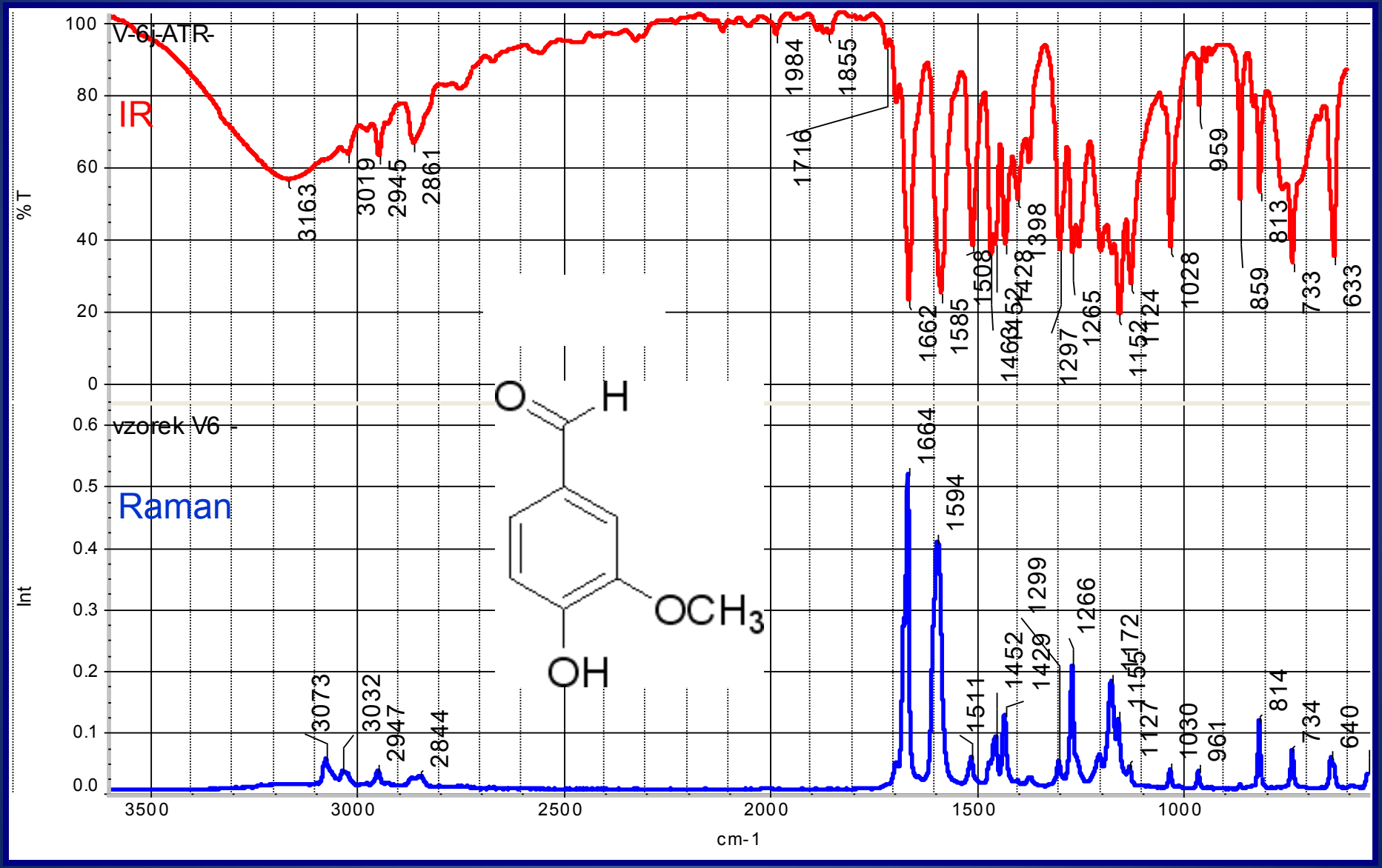
Identifikace drog



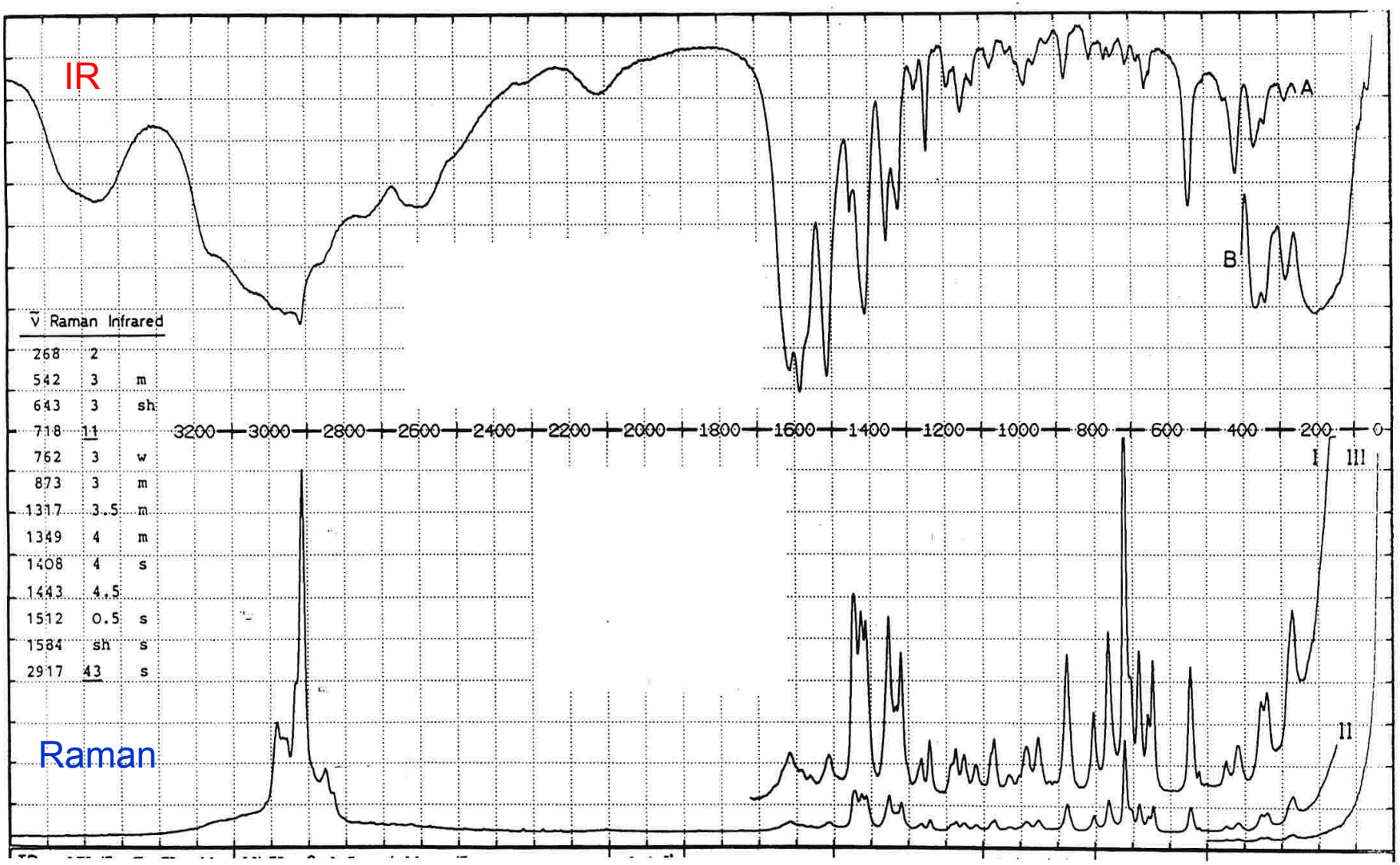
Identifikace léčiv



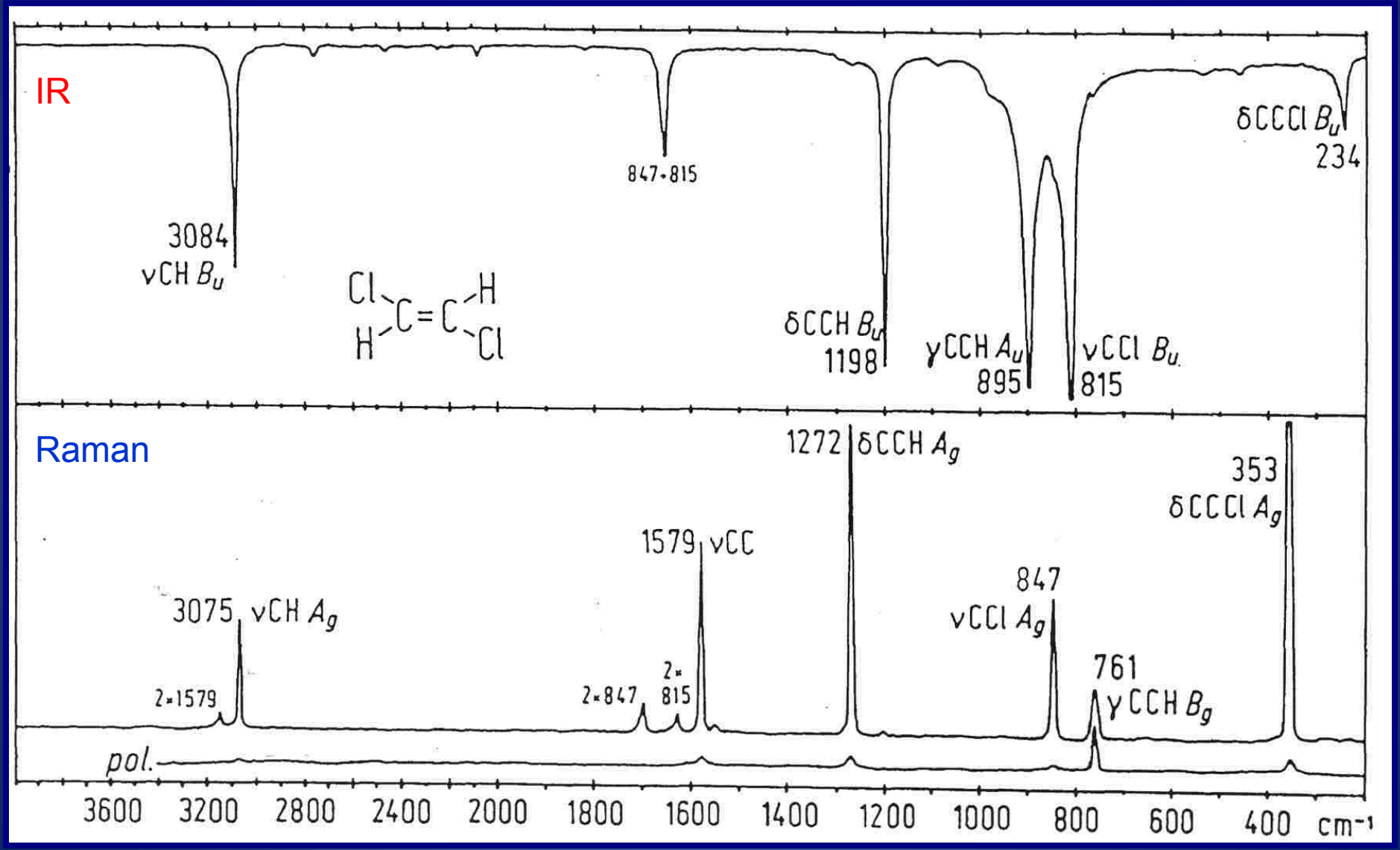
Rozdíly IČ a Ramanovy spektrometrie



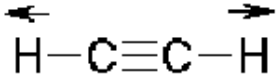
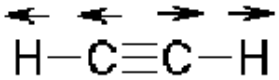
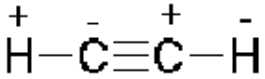
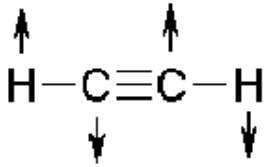
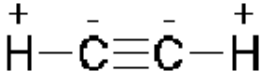
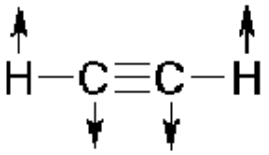
Rozdíly IČ a Ramanovy spektrometrie



Rozdíly IČ a Ramanovy spektrometrie



Rozdíly IČ a Ramanovy spektrometrie

Normal mode		Symmetry	Description	Activity	Wavenumber (cm^{-1})	
					C_2H_2	C_2D_2
	ν_1	Σ_g^+	CH stretching symmetric	R*p*	3373	2705
	ν_2	Σ_g^+	CC stretching (sym)	R*p*	1974	1765
	ν_3	Σ_u^+	CH stretching antisymmetric	IR	3295	2439
 	ν_4	Π_g	Deformation symmetric	R*dp*	613	512
 	ν_5	Π_u	Deformation antisymmetric	IR	730	539

Pohyb atomů v molekule
VIBRACE

INTERPRETACE VIBRAČNÍCH SPEKTER

- CHARAKTERISTICKÉ PÁSY FUNKČNÍCH SKUPIN
  - * zjištění typu skeletu a identifikace substituentů

- SPEKTRUM JAKO „OTISK PALCE“
  - * identifikace ČISTÉ LÁTKY
    - KNIHOVNY SPEKTER

Interpretace spekter

- srovnávání s databází
  - tištěné katalogy
  - elektronické knihovny
- tabulky charakteristických pásů
  - kombinované IR + Raman
- srovnání s vypočtenými spektry
  - kvantově chemické výpočty
    - ab initio, empirické

Normální vibrační módy a charakteristické vibrace funkčních skupin

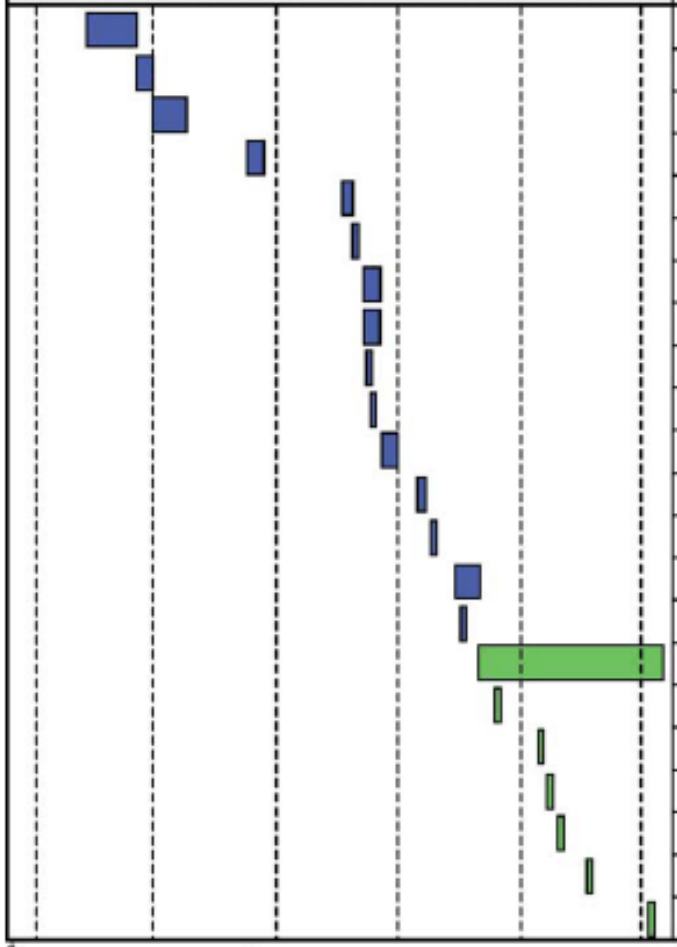
- **některé vibrační módy jsou lokalizovány jen v určité části molekuly**
  - **vibrace skupin s vodíky (lehké atomy)**
  - **vibrace násobných vazeb (odlišná síla vazby)**
  - **vibrace substituentů: $-NO_2$, $-SO_2$, -P=S, (-S-S-), -C=S -F, -Cl, -Br, -I**
  - **problém spřažení vibrací** (při srovnatelné hmotnosti atomů a srovnatelné síle vazeb)
  - **problém symetrie**
  - **problém vlivu okolí na sílu vazby** (posuny pásů)
  - **problém rigidity struktury, otázka konformerů**

Normální vibrační módy a charakteristické vibrace funkčních skupin

- **některé vibrační módy jsou lokalizovány jen v určité části molekuly**

- **vibrace skupin s vodíky**
  - **H**-C-C pod 3000 cm^{-1} $-CH_3$, $-CH_2$, -CH
  - **H**-C=C 3100 – 3000 cm^{-1} $-C=CH_2$, -C=CH-, Ar
  - **H**-C≡C 3340 – 3280 cm^{-1} (3333 cm^{-1} C_2H_2, 3310 cm^{-1} $HC\equiv C-C_5H_{11}$)
  - H-O, H-N, H-S, H-B

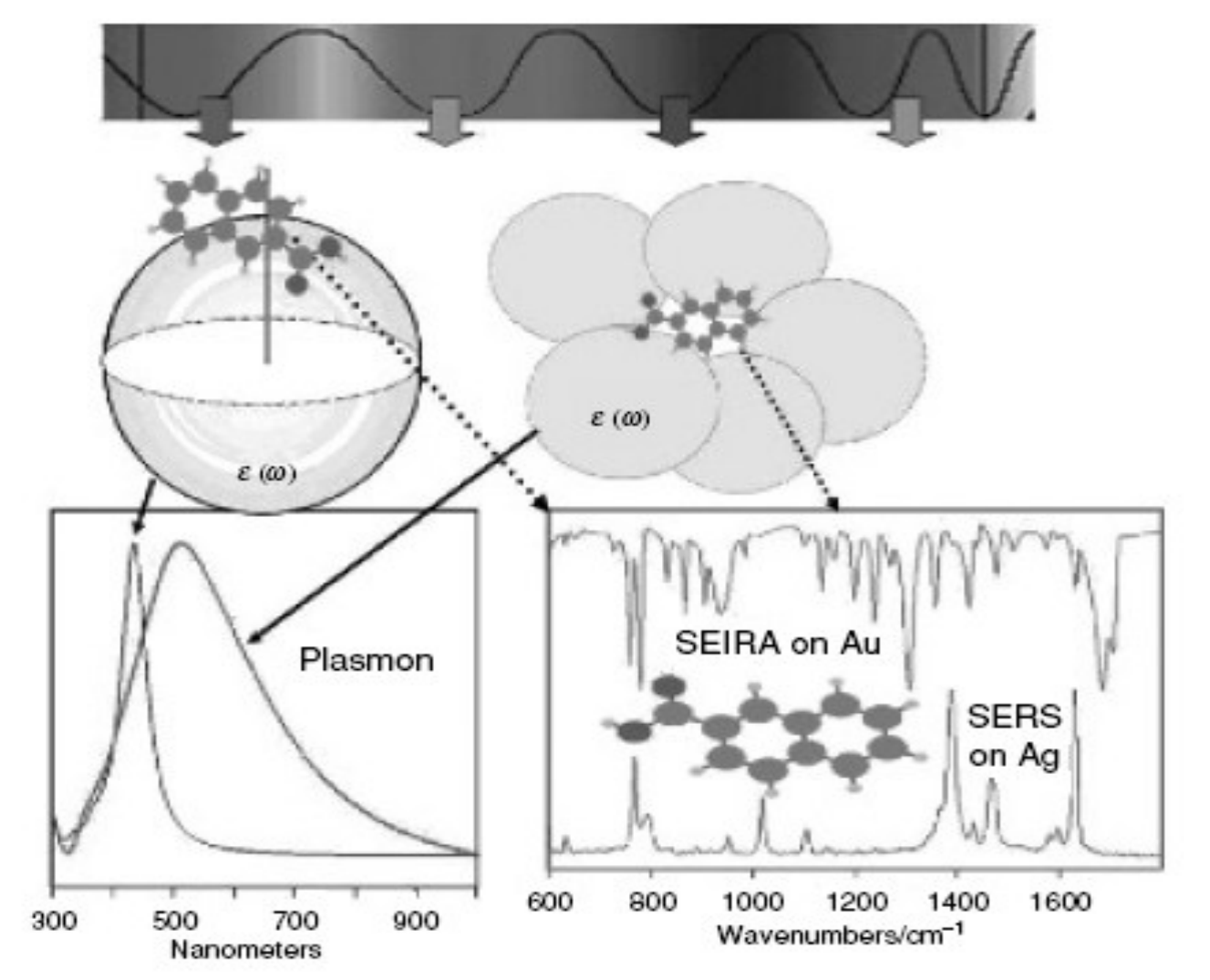
Stručná tabulka pro biologicky významné molekuly



Group Vibration	Intensity#		Description	Mainly observed in
	IR	Raman		
O-H stretch	vs	vw	Hydroxyl	Liquid phase
=C-H stretch	s-m	m	Unsaturated	Lipids
-C-H stretch	s-m	m	Saturated	Lipids
-C≡N stretch	m	s	Nitrile	
C=O stretch	s	m-w	Ester	Lipids, Amino Acid
C=O stretch	s	w-m	Carboxylic acid	Lipids, Amino Acid
C=O stretch	s	m-s	Amide I	Proteins
C=C stretch	m-w	s	Not conjugated	Lipids
C=C stretch	m	s	*Trans*	Lipids
C=C stretch	m	s	*Cis*	Lipids
N-H bending	s	w	Amide II	Proteins
C-H scissoring	m	m-w	Aliphatic -CH_2	Lipids
C-O stretch	s		Carboxylates	Amino Acids, Lipids
N-H bending	w-m	var	Amide III	Proteins
P=O stretch	vs	m-w	Phosphate ester	Lipids, Nucleic Acids
		Fingerprint from skeleton		
C-O stretch	s	m-w	Ether	Carbohydrates
Skeletal mode		m	α-(1→4) linkage	Starch
C-O-C skeletal	m-w	m-w	β-configuration	Glucose, galactose, mannose
C-O-C skeletal	m-w	m	α-configuration	
C-H rocking	w-m	vw	Aliphatic -CH_2	Lipids
Skeletal mode		vs		Starch

s = strong, m = medium, vs = very strong, vw = very weak

Povrchem zesílené spektroskopie



SERS

- **surface enhanced** – povrchem zesílený
  - **zesílený signál od specií adsorbovaných na povrchu stříbra, zlata a mědi – faktor zesílení – více než 10^4** (lokálně i více než 10^{12}) - až k detekci jednotlivých molekul
  - **zesílení závisí na morfologii povrchu – "hrubý (nerovný) povrch" – nanostrukturovaný, nanočástice**

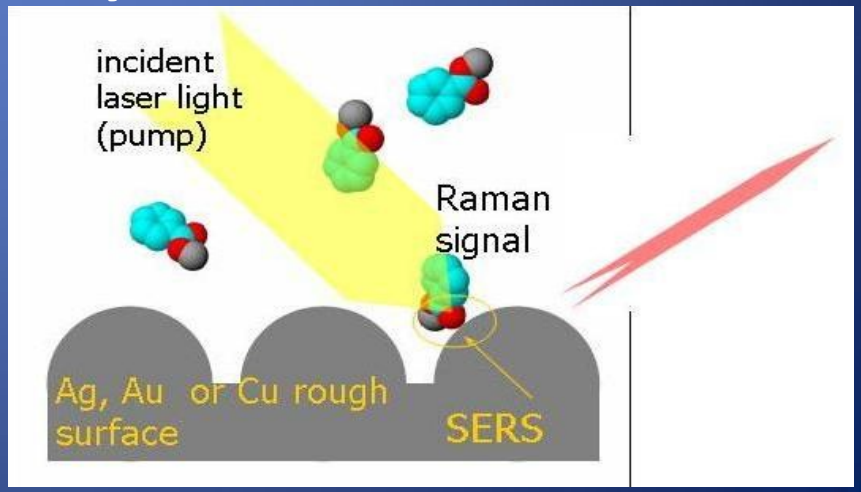
$$\vec{P} = \alpha \vec{E}$$

  - **dva mechanismy – elektromagnetický (povrchová plasmonová resonance), chemický – chemisorpce, komplexy s přenosem náboje**
  - **SERS-aktivní substráty – zdrsněné elektrody, koloidní částice, ostrůvkové filmy, mezifázové filmy …**

SERS

A new field is born, apparently in full adulthood, and complete with a name. Such was the case with the Mössbauer effect and with polywater, and so, too, was the case with SERS. The first resulted in Nobel Prize, the second was shown to be spurious; SERS, I believe, has settled in the territory between.
M. Moskovits

- **SERS** (surface-enhanced Raman scattering)
- **zesílený signál od specií adsorbovaných na povrchu stříbra, zlata a mědi – faktor zesílení – více než 10^4** (lokálně i více než 10^{12}) - až k detekci jednotlivých molekul
- **zesílení závisí na morfologii povrchu – "hrubý (nerovný) povrch" – nanostrukturovaný, nanočástice**

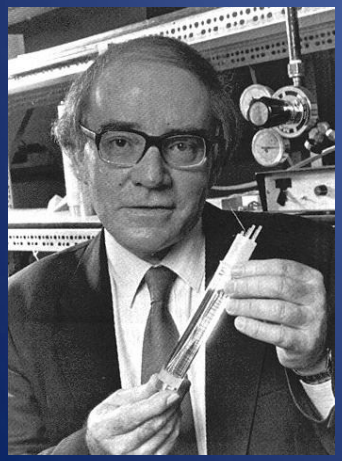


SERS

- 70. léta – objev povrchem zesíleného Ramanova rozptylu (SERS)

Fleischmann M., Hendra P.J., McQuillan A.J.; Chem. Phys. Lett. **26**, (1974) 163.

- pozorování velmi intenzivního spektra pyridinu na stříbrné elektrodě – vysvětleno nárůstem plochy povrchu elektrody po jejím zdrsnění



Fleischmann Martin

- nar. 29.3. 1927 - Karlovy Vary
- od 1938 v Anglii
- 1973-74 - prezident - the International Society of Electrochemistry

SERS

doi:10.1016/0009-2614(74)85388-1
 Cited By in Scopus (1155)

Raman spectra of pyridine adsorbed at a silver electrode

M. Fleischmann[a], P.J. Hendra[a] and A.J. McQuillan[a]
[a]Department of Chemistry, The University, Southampton SO9 5NH, UK
Received 27 February 1974.
Available online 20 November 2001.

Abstract
Raman spectroscopy has been employed for the first time to study the role of adsorption at electrodes. It has been possible to distinguish two types of pyridine adsorption at a silver electrode. The variation in intensity and frequency of some of the bands with potential in the region of the point of zero charge has given further evidence as to the structure of the electrical double layer; it is shown that the interaction of adsorbed pyridine and water must be taken into account.
Chemical Physics Letters
Volume 26, Issue 2, 15 May 1974, Pages 163-166

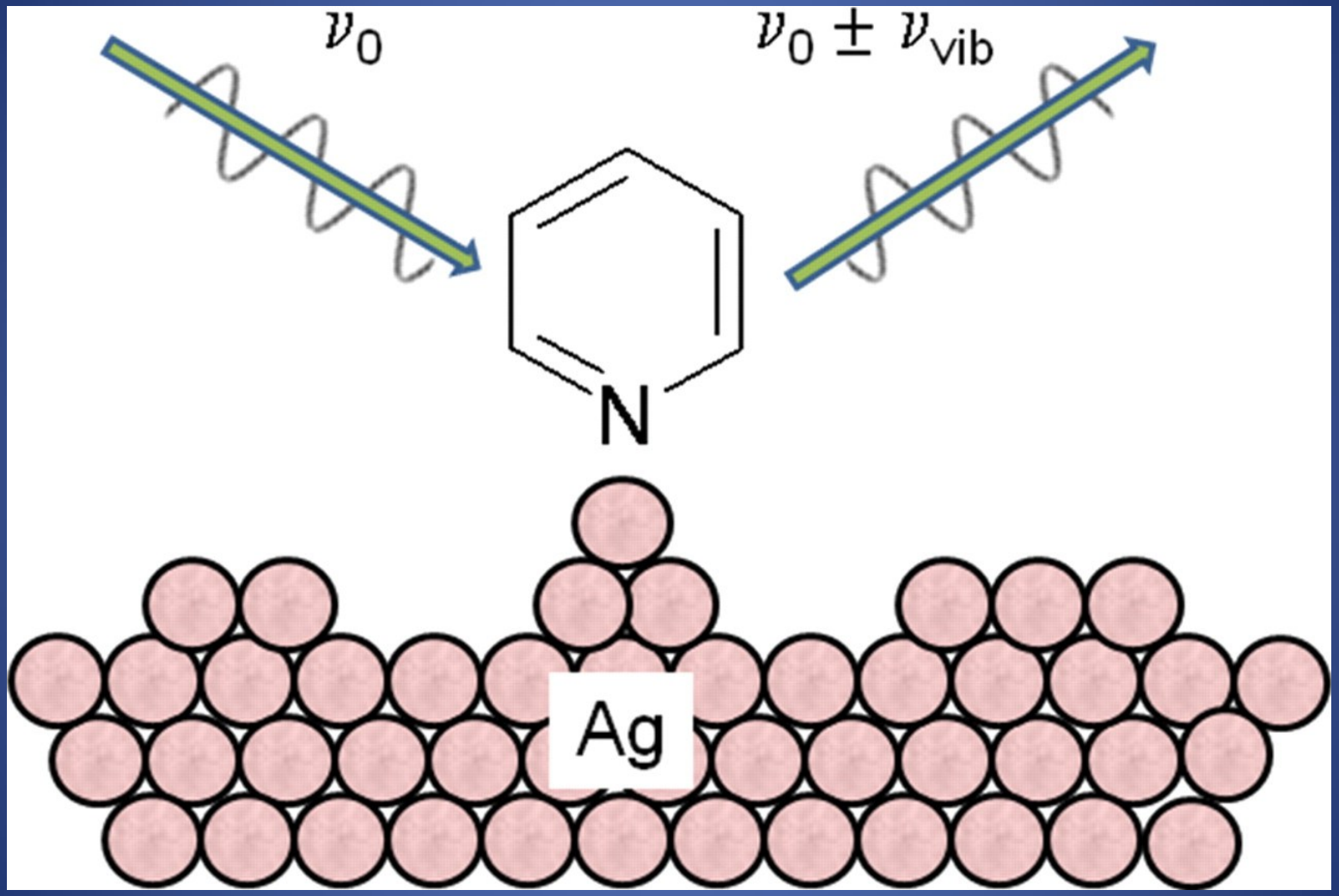
SERS

- 70. léta – objev povrchem zesíleného Ramanova rozptylu (SERS)

1977 - rozpoznání nového fyzikálního jevu – zesílení Ramanova rozptylu díky zesílení elektromagnetického pole v blízkosti povrchu stříbra

- Jeanmaire D.L., van Duyne R.P.; J. Electroanal. Chem. Interfacial Electrochem. **84**, (1997) 1.
  - Ag elektroda, pyridin, aminy, zesílení el-mg. pole na elektrické dvojvrstvě
- Albrecht M.G., Creighton J.A.; J. Am. Chem. Soc. **99**, (1977) 5215.
  - Ag elektroda, pyridin, resonance – CT adsorbát-povrch

A schematic representation of a SERS experiment with pyridine adsorbed on silver, showing the incident laser and Raman scattered light, the intensities of which are both influenced by the enhanced field at the silver surface resulting from surface plasmon excitations.

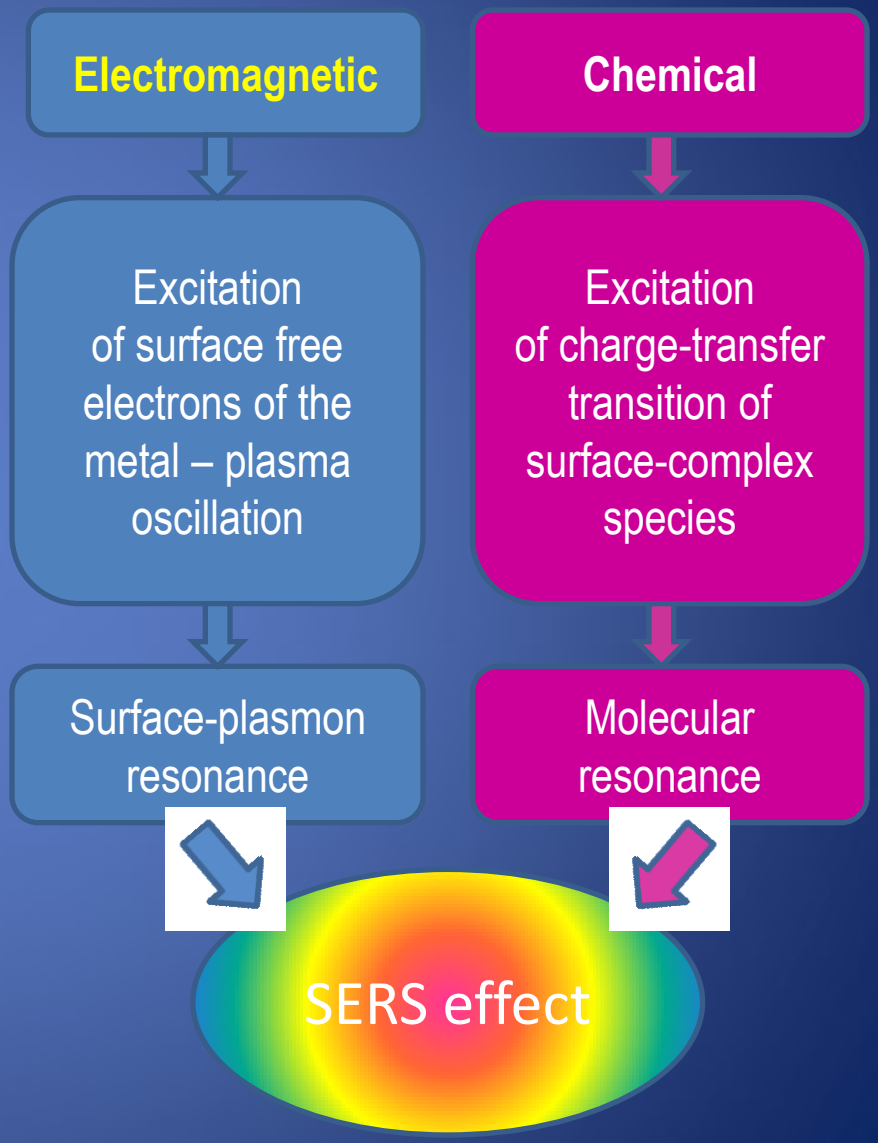


McQuillan A J Notes Rec. R. Soc. 2009;63:105-109

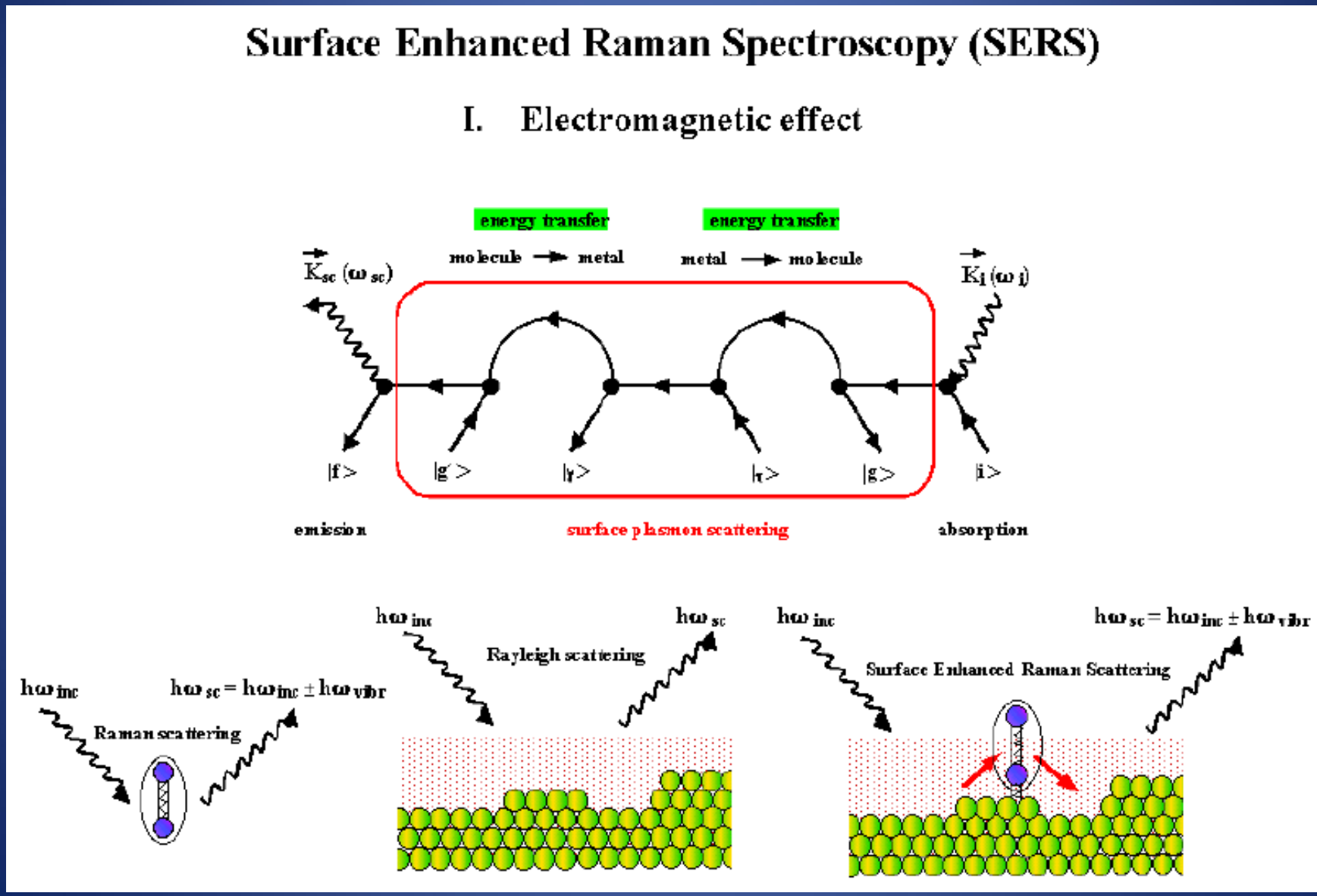
NOTES & RECORDS OF THE ROYAL SOCIETY

SERS Spectroscopy

- giant enhancement of Raman signal
- two mechanisms involved
  - **electromagnetic** - long range, depends on metal-substrate properties (surface plasmons are involved) – coin metals – Au, Ag, Cu
  - **chemical** - local, molecular structure plays an important role (formation of surface complex)



Povrchem zesílený Ramanův rozptyl



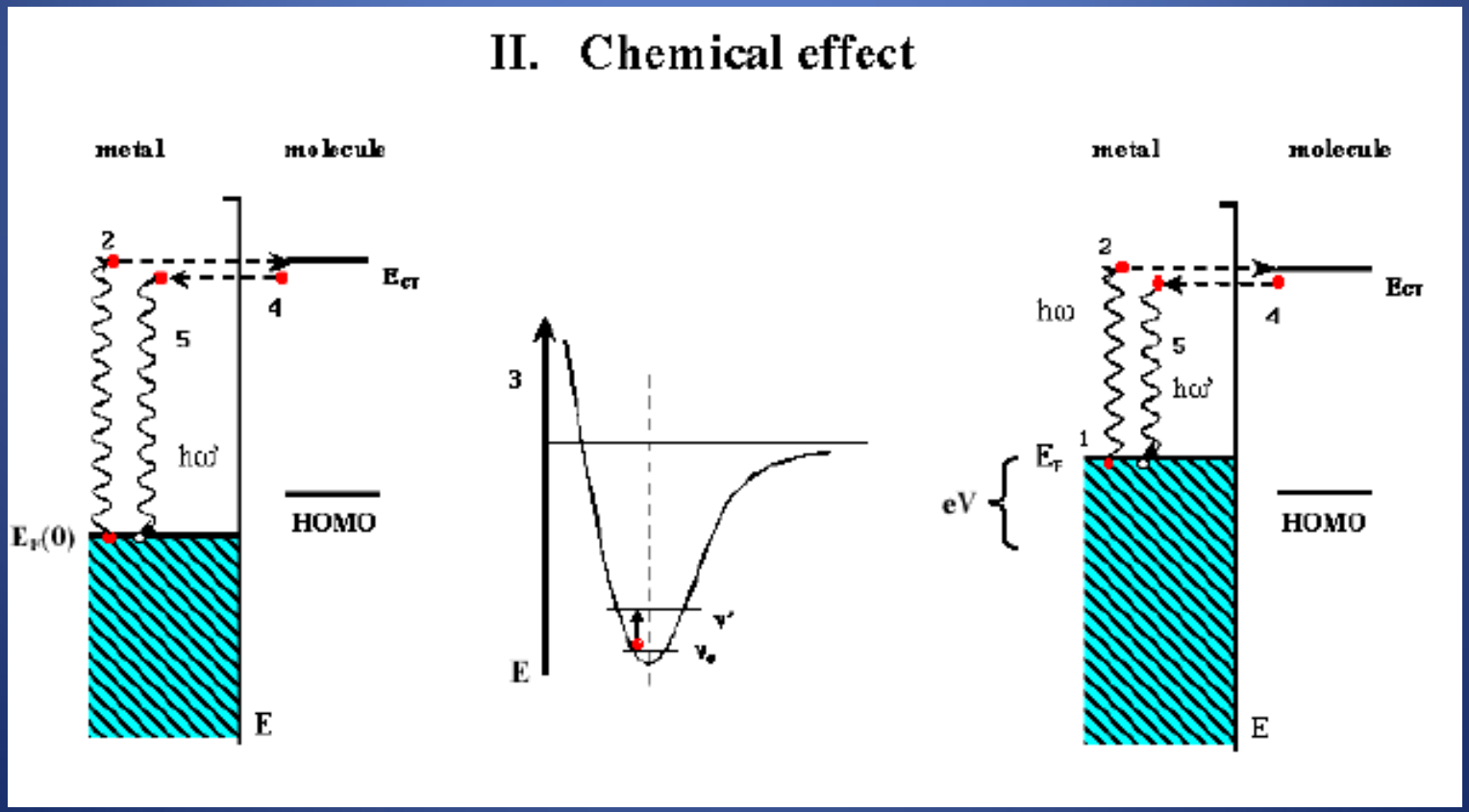
SERS

- Chemický mechanismus
- **vybuzení „molekulového“ rezonančního Ramanova rozptylu pro povrchový komplex – přenos náboje mezi adsorbátem a povrchem kovu**

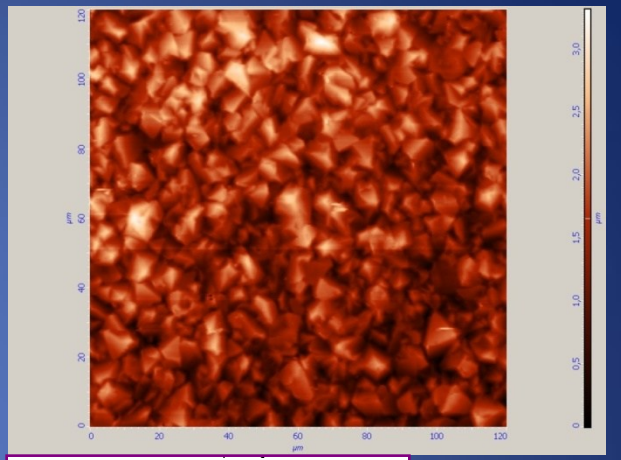
- **Modely**
  - **HOMO – LUMO – analogická koordinační sloučenina**
  - **zahrnutí několika atomů – (nabitého) klastru**
  - **vliv „povrchového“ potenciálu**

Povrchem zesílený Ramanův rozptyl

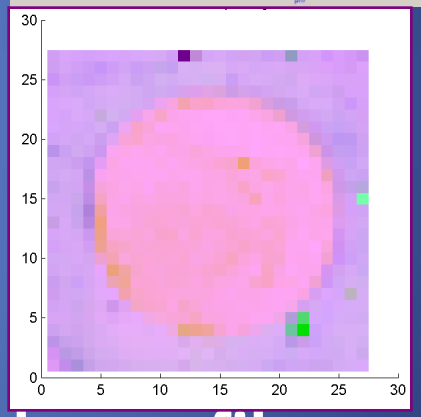
– chemický mechanismus – efekt přenosu náboje (CT), povrchový komplex – analogie k RR, otázka potenciálu (elektrody)



Povrchem zesílený Ramanův rozptyl



– **Intenzita signálu**
  - **Vliv druhu kovu**
  - **Vliv morfologie povrchu**



  - **Vliv pokrytí adsorbátem**
    – **orientace adsorbátu vůči povrchu**

  - **vliv excitační vlnové délky (otázka profilu povrchové plasmonové rezonance a molekulární rezonance)**

Povrchem zesílený Ramanův rozptyl

– **specifická výběrová pravidla** –
spektra odlišná oproti normálnímu Ramanově spektru, příp. oproti rezonančnímu Ramanově spektru

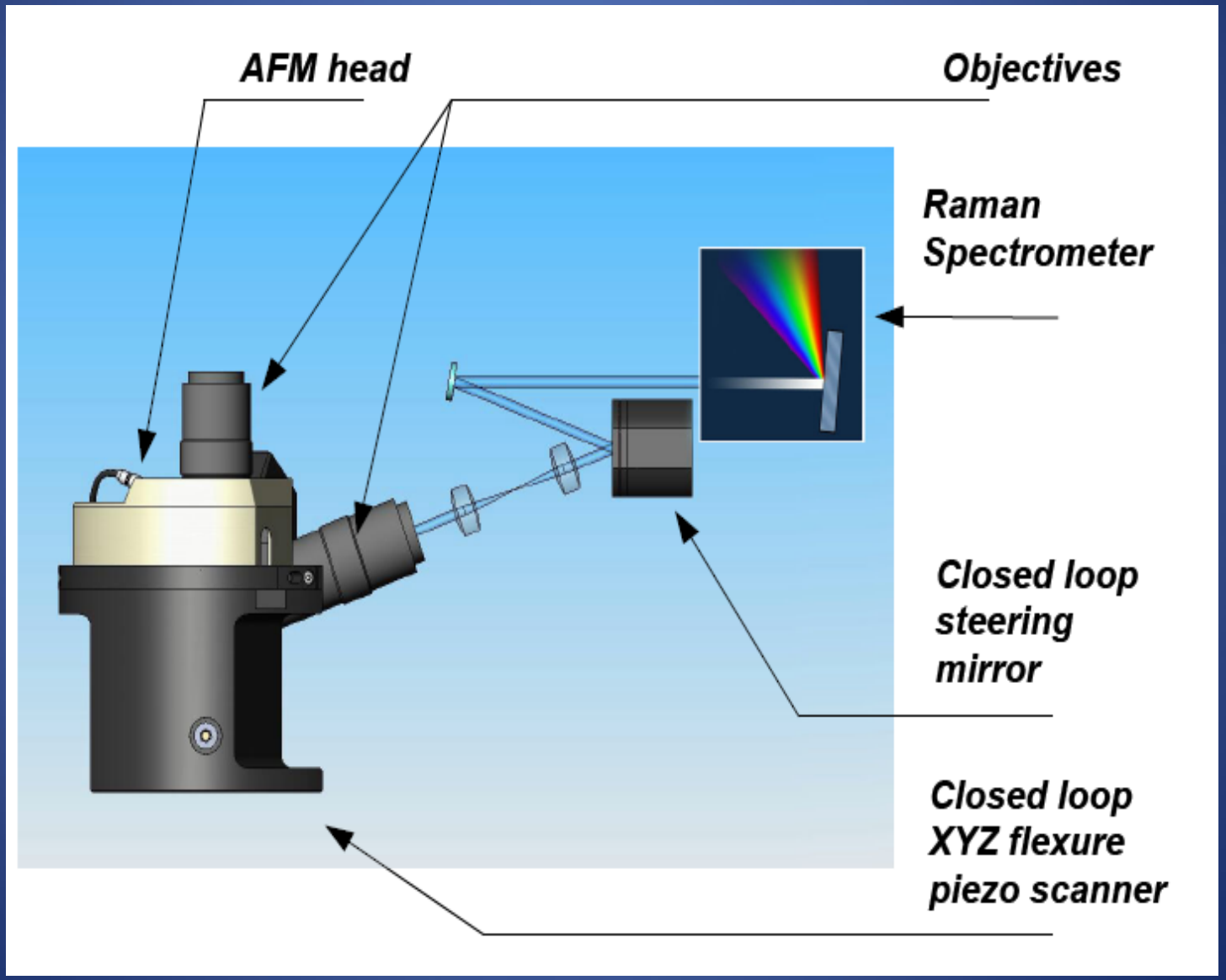
❑ elektromagnetický mechanismus

– módy kolmé k povrchu více zesíleny než módy paralelně orientované s povrchem

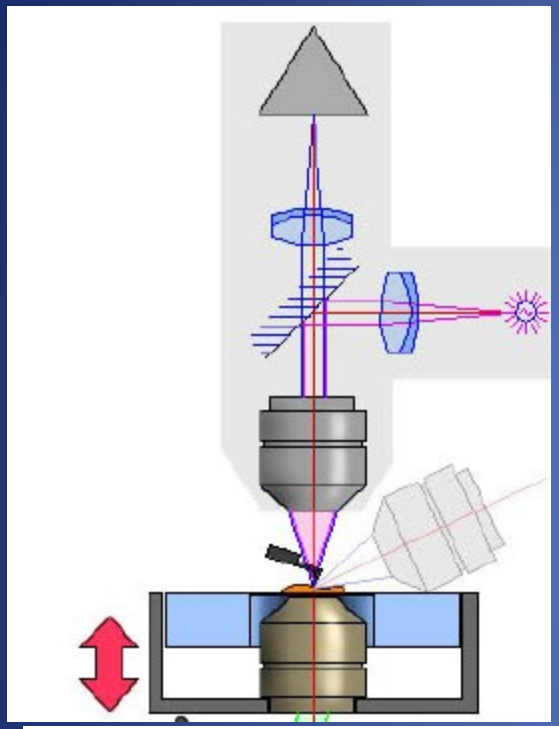
❑ chemický mechanismus

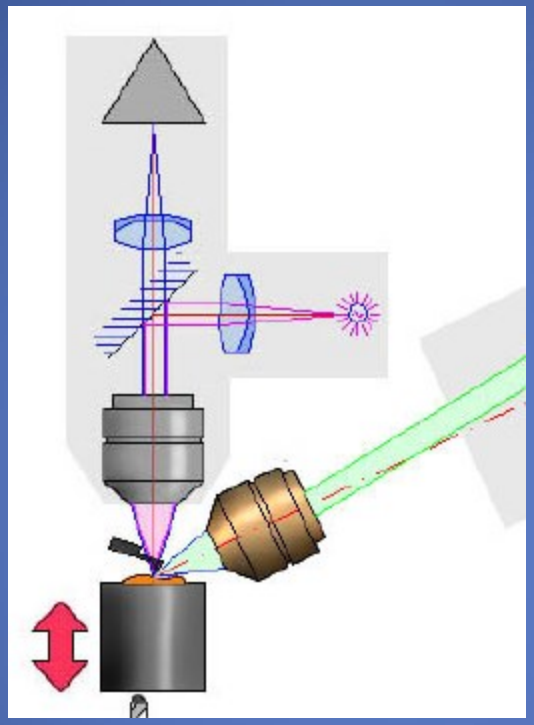
– změna symetrie „povrchového komplexu“

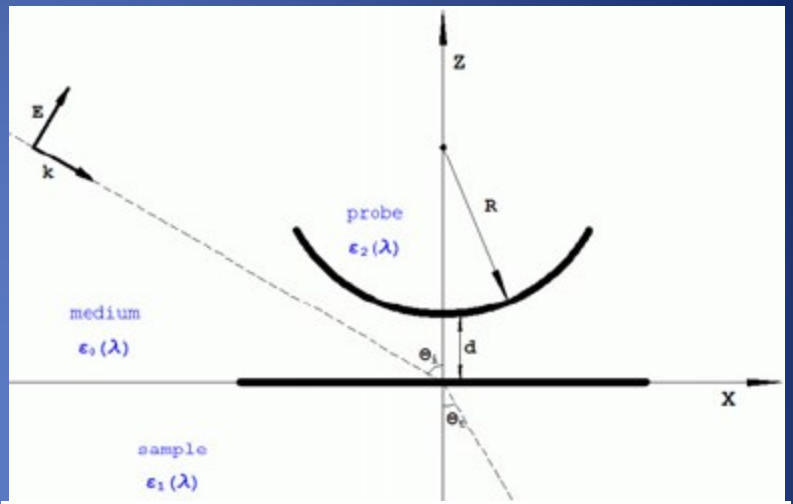
Instrumentace
- integrované AFM + TERS

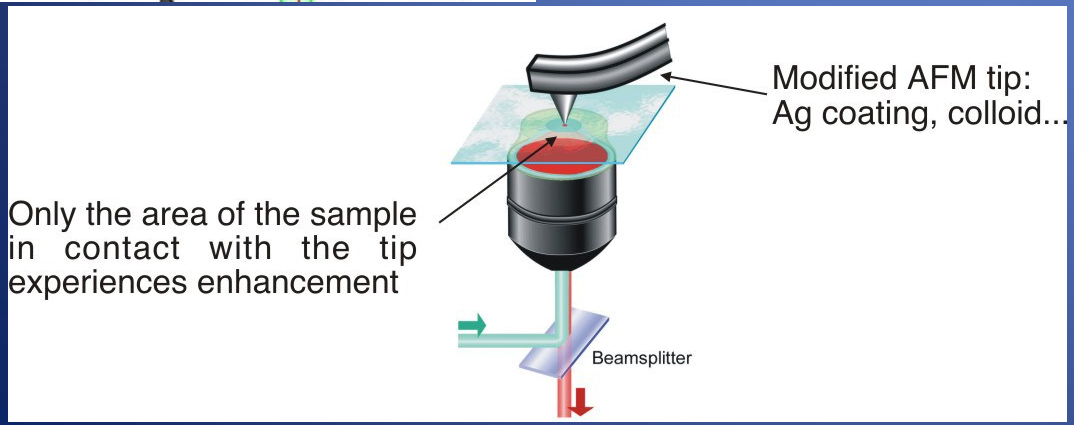


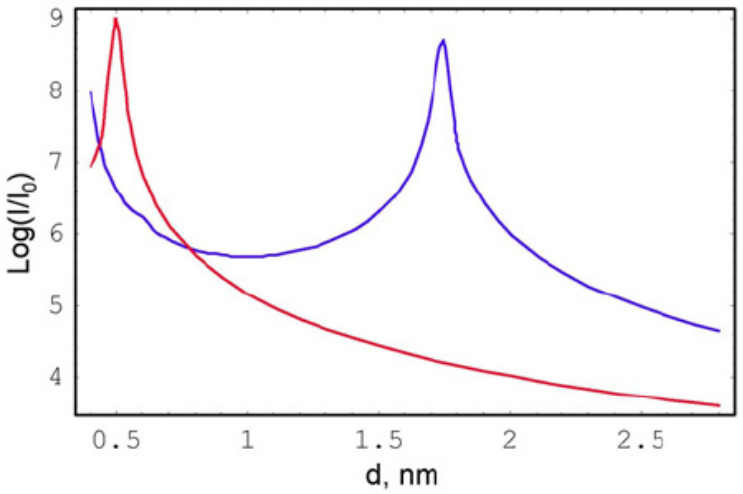
Instrumentace - integrované AFM + TERS
dva optické porty







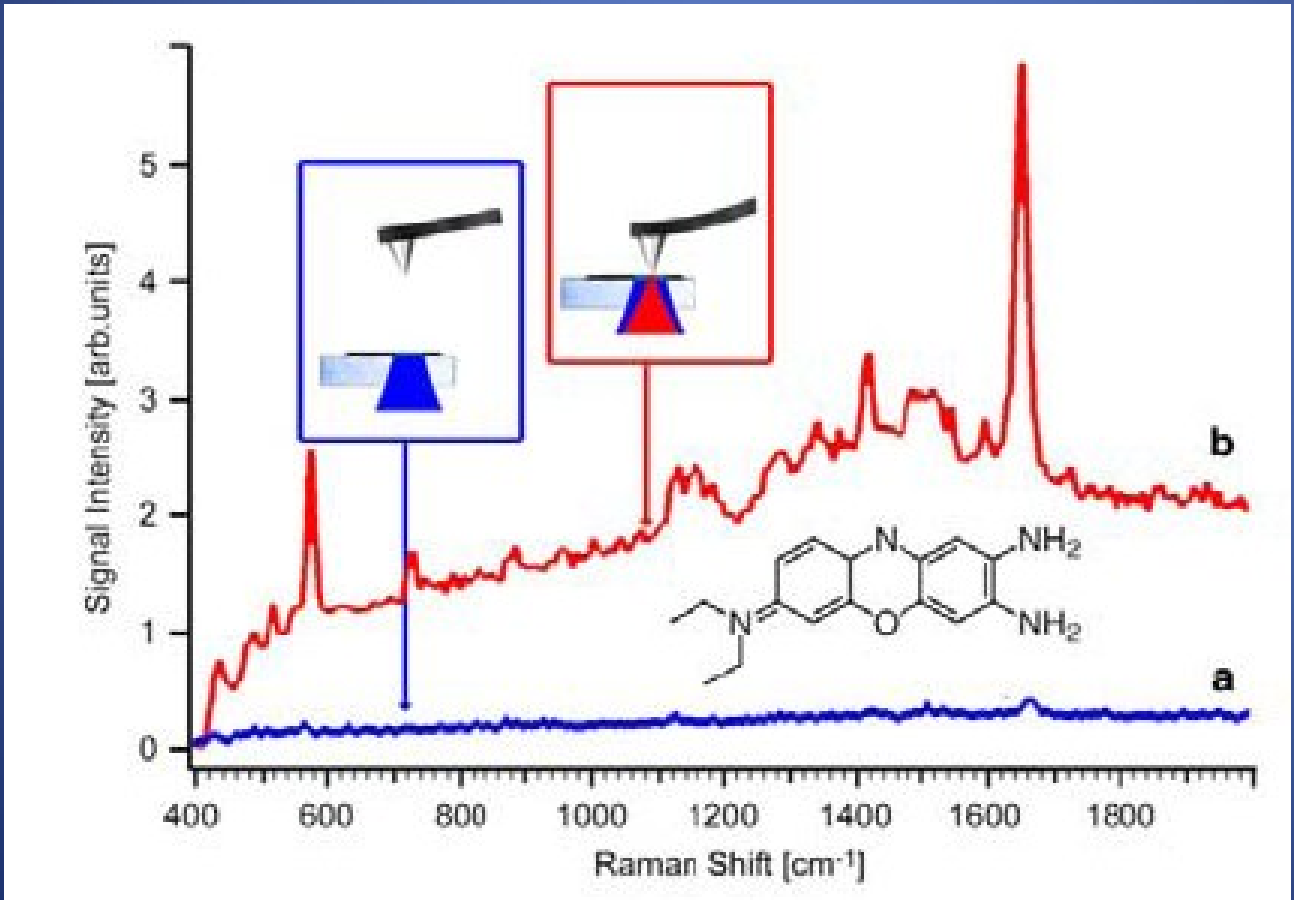




Local field intensity dependence (logarithmic scale) on the probe-sample distance d.
Here R=100 nm, X=0 nm, Y=0 nm, Z=1 nm; light wavelength: 653 nm and 775 nm.

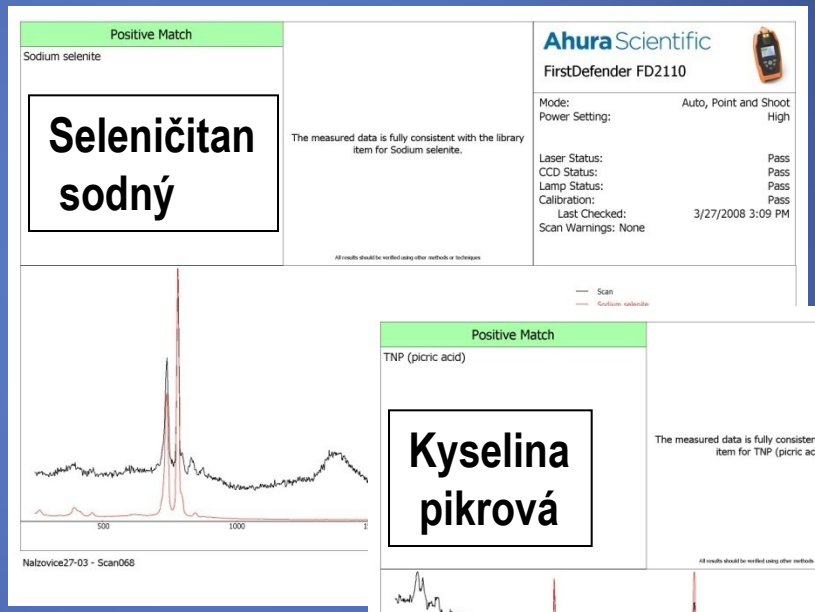
Kombinace

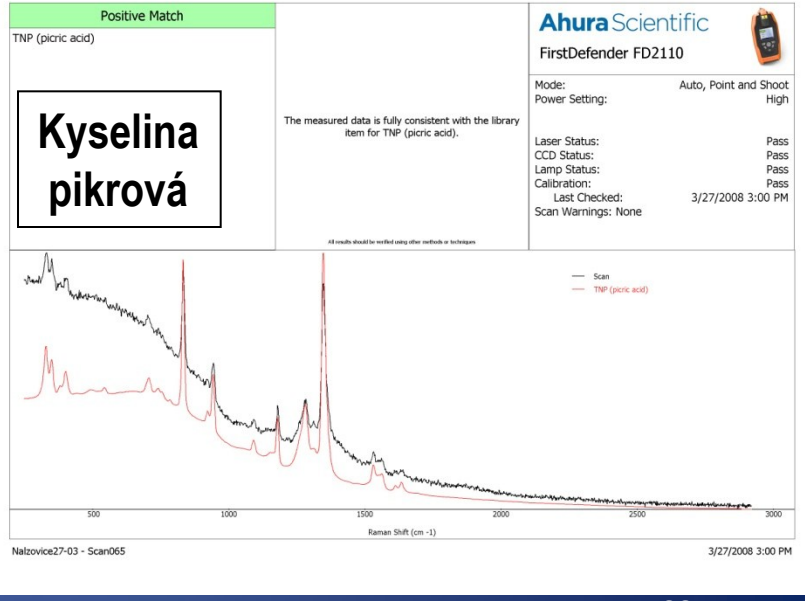
– TERS, AFM



Terénní měření

- Přenosný dispersní Ramanův spektrometr – AHURA
- Nalžovice – areál bývalého statku





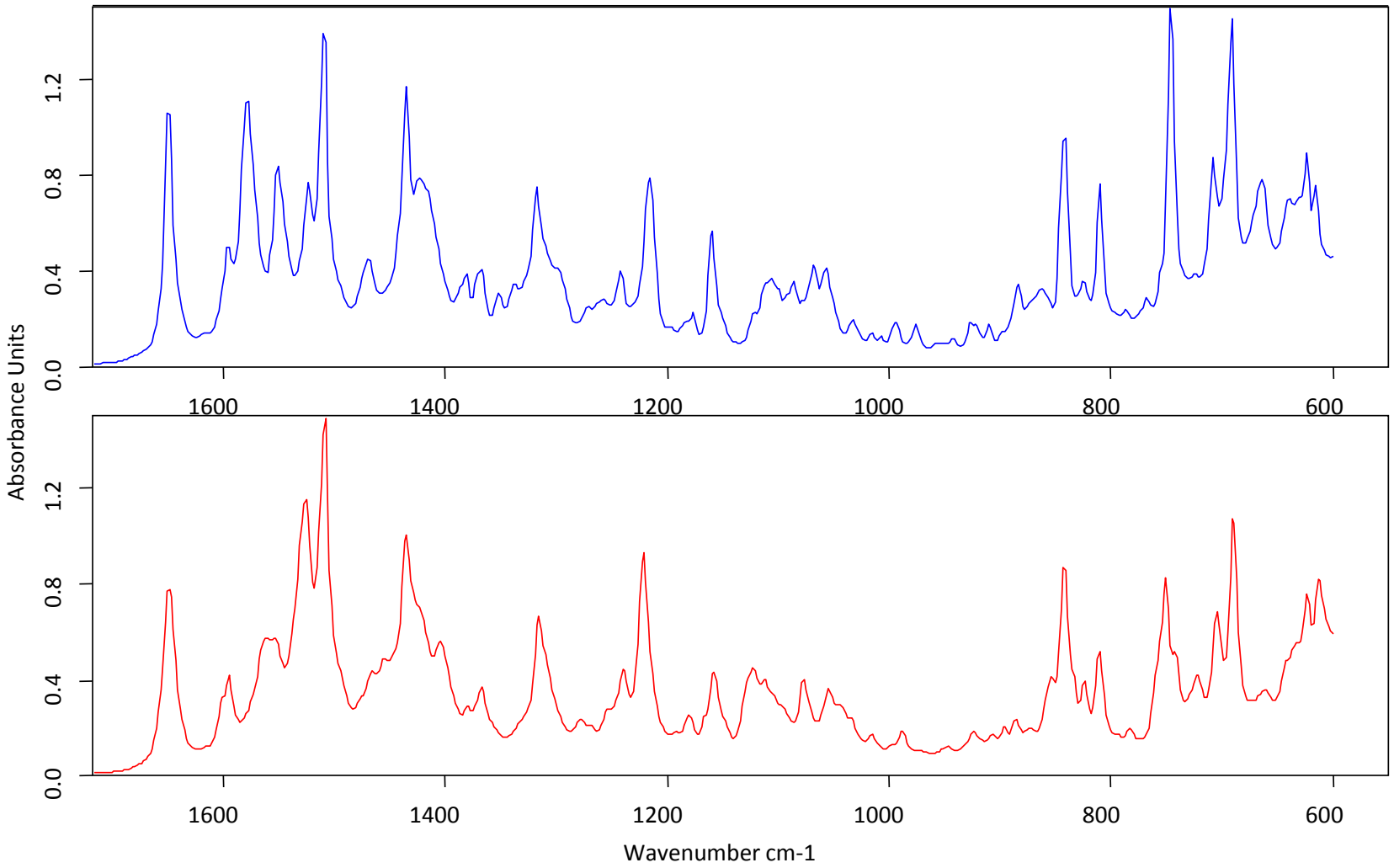
Terénní měření

- Přenosný dispersní Ramanův spektrometr – AHURA

Léčiva

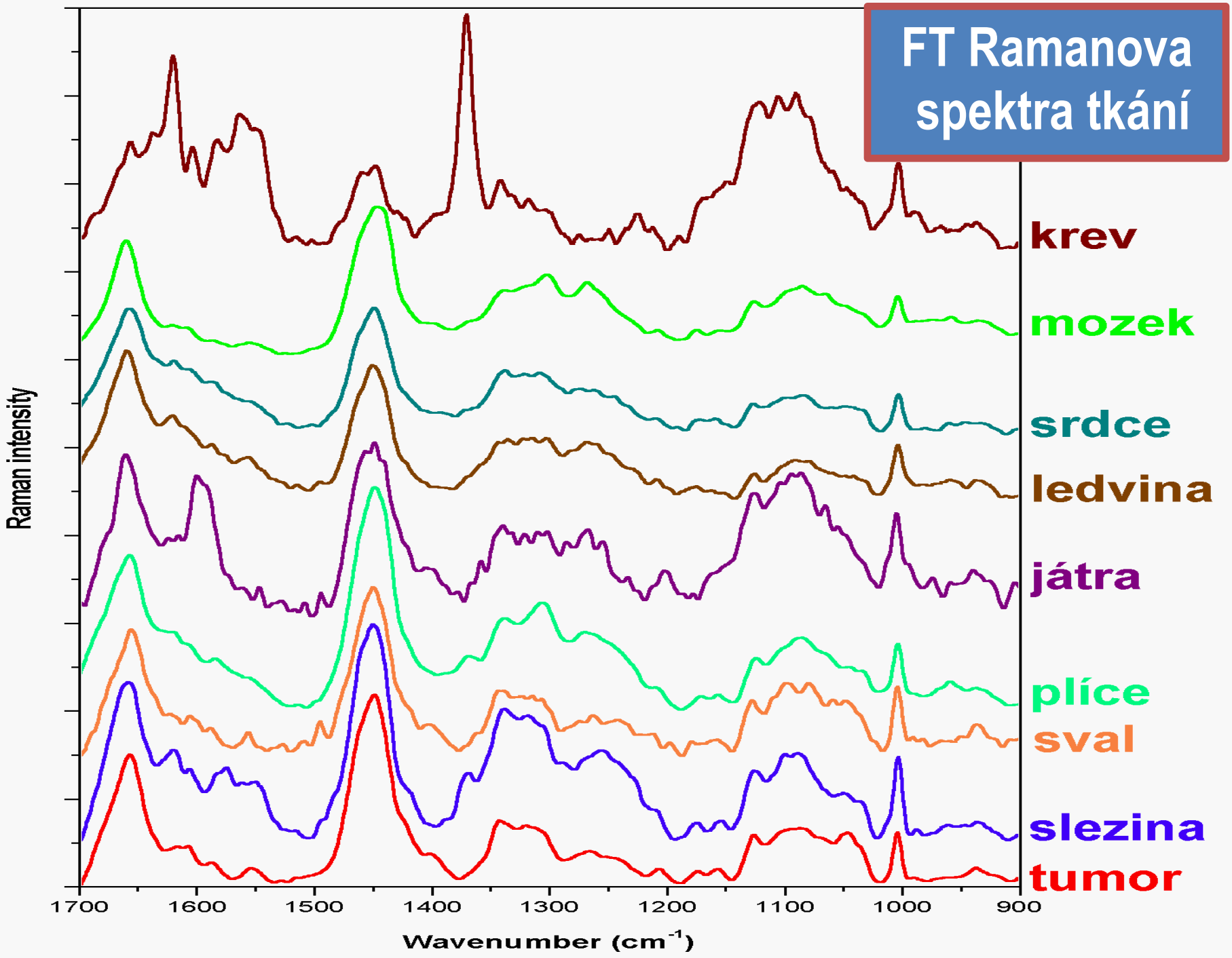
Ramanova spektroskopie – přínos pro polymorfní studie

- Studium přesycených roztoků a agregátů – identifikace povahy polymorfu před jeho vznikem identifikací intermolekulárních interakcí
- Studium přechodů anebo desolvatace za proměnné teploty (ve spojení s faktorovou analýzou)
- Vibrace mřížky u nízkých vlnočtů



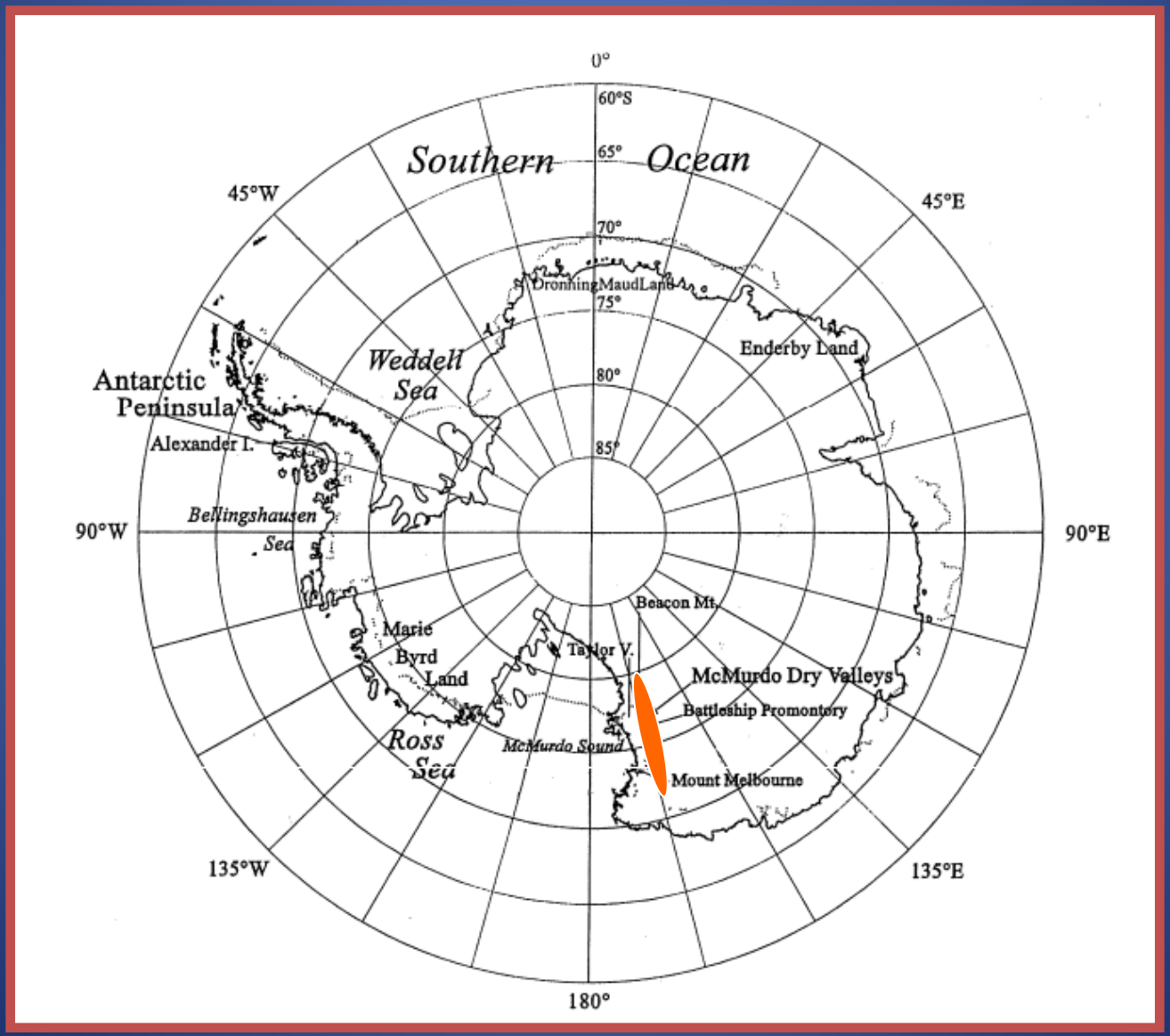
Srovnání FT-Ramanových spekter dvou polymorfů

FT Ramanova spektra tkání



Život v extrémních podmínkách

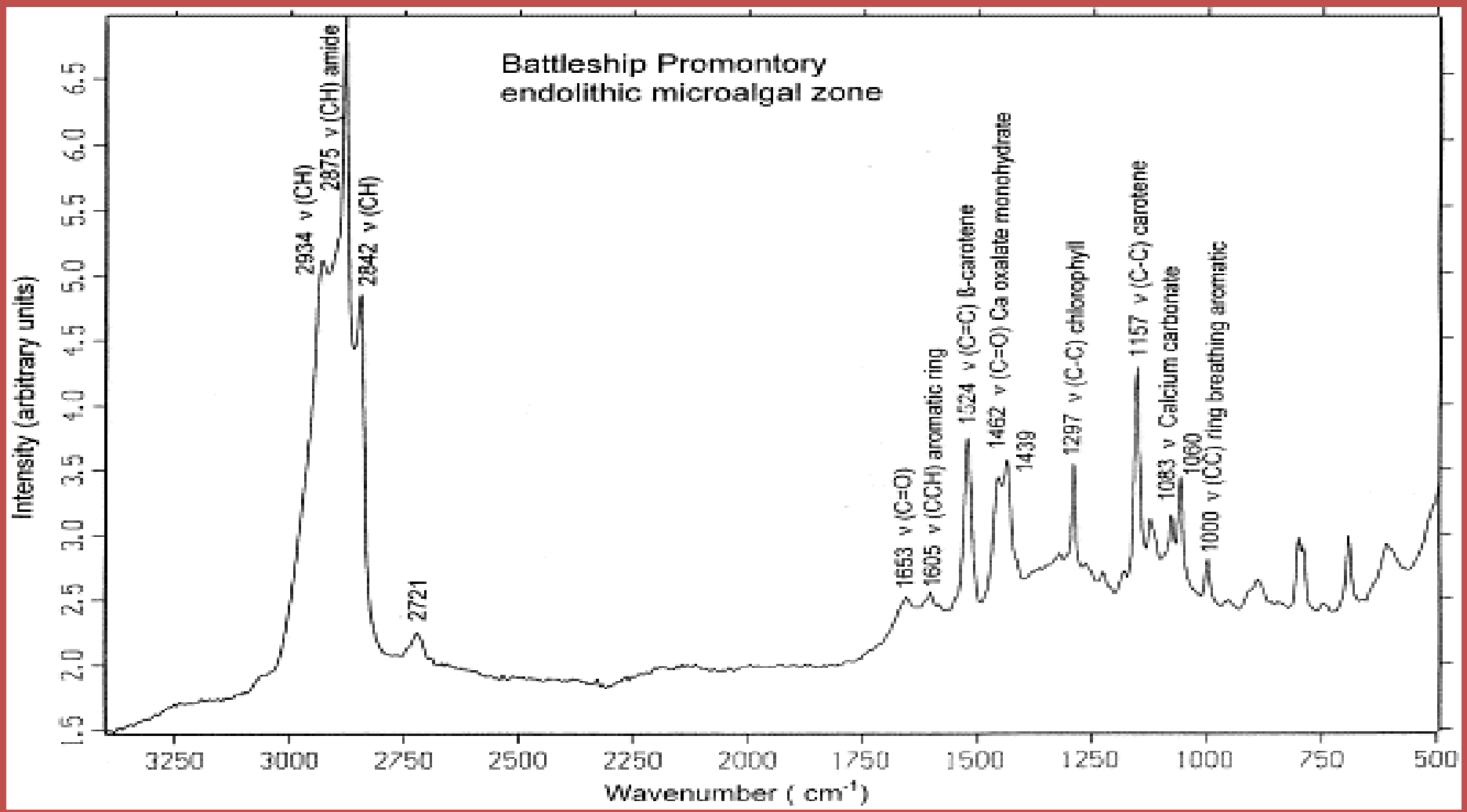
• ANTARKTIDA



Život v extrémních podmínkách

FT Ramanovo spektrum - endolithické společenství

D.D. Wynn-Williams, H.G.M. Edwards
Planetary and Space Science **48** (2000) 1065.



Život na Marsu ?

- **DETEKCE BIOMOLEKUL**
  - pigmenty a chinoidní látky chránící před UV zářením
  - pigmenty potřebné pro fotosyntézu
  - šťavelany a další látky typické pro primitivní organismy v Antarktidě

- **VYUŽITÍ Ramanovy spektroskopie**
  - snadná detekce a identifikace pigmentů
  - snadná detekce a identifikace chinoidních látek
  - možnost detekce dalších organických i biogenních anorganických látek

Život na Marsu ?

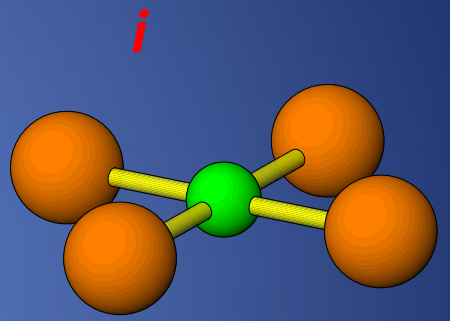
- **Metodická příprava**
  - ➢ **vývoj spektrometru MaRS** (microscope and Raman spectrometer)
  - ↳ **miniaturní, lehký, a přitom robustní**
  - ↳ **prototyp - Montana State University**

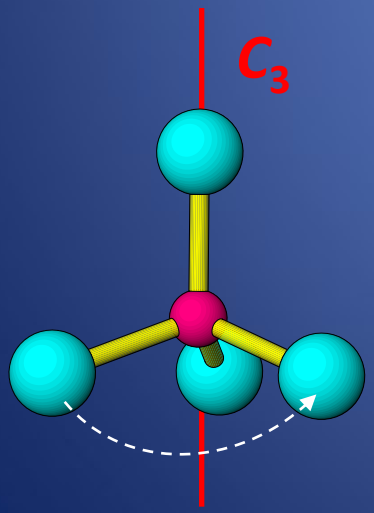
D.L. Dickensheets, D.D. Wynn-Williams, H.G.M. Edwards, C. Schoen, C. Crowder, E.M. Newton,
Journal of Raman Spectroscopy **31** (2000) 633.

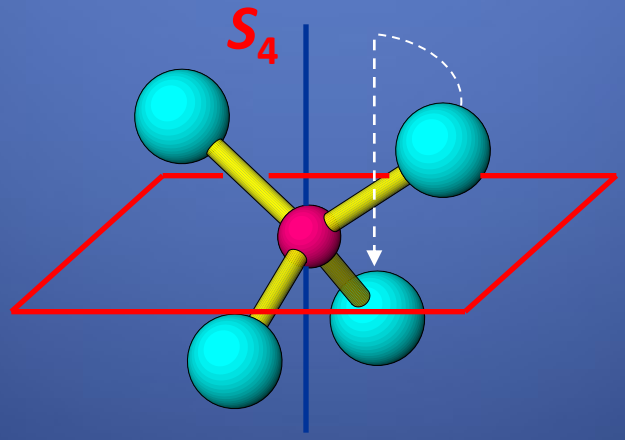
Symetrie molekul

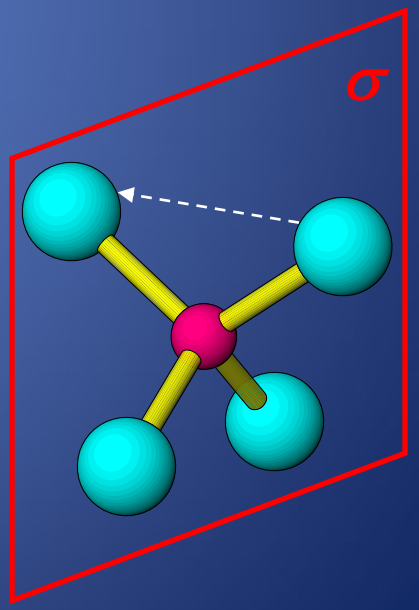
Prvky / *operace* symetrie

- **identita – *E***
- **střed symetrie / *inverze* – *i***
- ***n*-četná osa symetrie / *rotace* – C_n**
- **rovina symetrie / *zrcadlení, reflexe* – σ**
- ***n*-četná rotačně-reflexní osa – S_n**



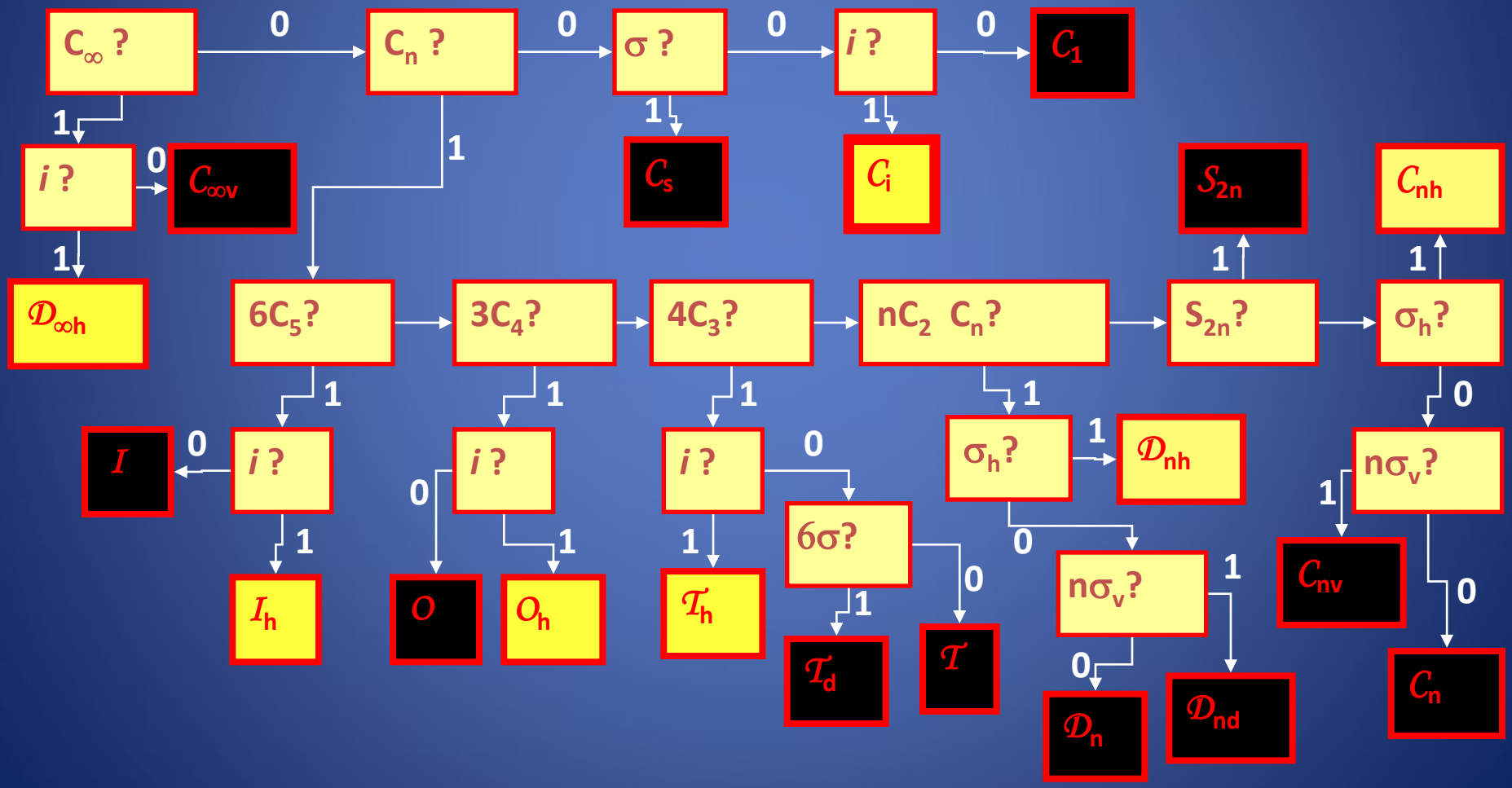






BODOVÉ GRUPY SYMETRIE

Množiny operací symetrie – grupy – grupové postuláty



Bodové grupy a tabulky charakterů

- **Operace symetrie** podle **prvků** symetrie, kdy alespoň jeden bod je nezměněn (nepřesunut).
- Klasifikace molekul – bodově grupové reprezentace molekul.
- Užitečné informace o bodové grupě – **tabulky charakterů**.
- V horním levém rohu tabulky charakterů - symbol bodové grupy.
- Zbytek 1.řádku - operace symetrie příslušející dané bodové grupě.
- Písmena v levém sloupci - symboly irreducibilních reprezentací grupy.
- Čísla v tabulce - charaktery.
- Vpravo od čísel - symboly ***x***, ***y***, a ***z*** (osy), R_x, R_y, a R_z (rotace podél os) a x^2, ***xy***, ***xz***, y^2, ***yz***, z^2 atd. (“produkty” os)

C_i	*E*	*i*	_	_
A_g	1	1	R_x; R_y; R_z	x^2; y^2; z^2; xy; xz; yz
A_u	1	-1	x; y; z	

Bodové grupy a tabulky charakterů

- Fundamentální přechod je **aktivní v IČ spektru** (dává vznik absorpčnímu pásu), jestliže příslušný normální mód náleží stejné reprezentaci jako kterákoli z kartézských souřadnic (***x***, ***y***, a ***z***).
- Fundamentální přechod je **aktivní v Ramanově spektru**, jestliže příslušný normální mód náleží stejné reprezentaci jako jedna či více komponent tenzoru polarizovatelnosti (***x²***, ***xy***, ***xz***, ***y²***, ***yz*** a ***z²***).

C_i	*E*	*i*	_	_
A_g	1	1	R_x; R_y; R_z	***x²; y²; z²; xy; xz; yz***
A_u	1	-1	***x; y; z***	

Grupa C_i

C_i	*E*	*i*	_	_
A_g	1	1	R_x; R_y; R_z	x^2; y^2; z^2; *xy; xz; yz*
A_u	1	-1	*x; y; z*	

Grupa C_i je podgrupou všech bodových grup,

které obsahují operaci **inverze**

⇒ reprezentace ***g*** (symetrické vzhledem k inverzi)

⇒ ***u*** (antisymetrické vzhledem k inverzi)

(-x,-y,-z) ← (x,y,z) **dipólový moment se transformuje podle *u***

(x^2,y^2,z^2,xy,xz,yz) ← (x^2,y^2,z^2,xy,xz,yz) **polarizovatelnost se transformuje podle *g***

- http://www.chemistry.nmsu.edu/studntres/chem639/cgi-bin/group1.cgi

Irreducibilní reprezentace

A – symetrické vzhledem k C_n
B – antisymetrické vzhledem k C_n
E – 2x degenerované
T – 3x degenerované

$_1$ – symetrické vzhledem k σ_v nebo C_2

$_2$ – antisymetrické vzhledem k σ_v nebo C_2

$_g$ – symetrické vzhledem k *i*
$_u$ – antisymetrické vzhledem k *i*

otázka počtu stupňů volnosti

jakoukoliv reducibilní reprezentaci (molekuly) lze rozložit na lineární kombinaci ireducibilních

$$n_\Gamma = \frac{1}{h}\sum_g n_g \chi_R \cdot \chi_\Gamma$$

n_Γ četnost dané ireducibilní reprezentace

h řád grupy

n_g počet operací v dané třídě

χ_R charakter operace symetrie v reduc. repr.

χ_Γ charakter operace symetrie v ireduc. repr.

Grupa C_{2v}

C_{2v}	E	C_2	$\sigma_v(xz)$	$\sigma_v(yz)$	_	_
A_1	1	1	1	1	z	x^2; y^2; z^2
A_2	1	1	-1	-1	R_z	xy
B_1	1	-1	1	-1	x; R_y	xz
B_2	1	-1	-1	1	y; R_x	yz

Bodová grupa C_{2v}

– všechny symetrické reprezentace normálních módů: A_1, A_2, B_1 a B_2, dovolené pro Ramanův rozptyl

– žádné normální módy nejsou zakázané z pohledu Ramanova rozptylu

– příklad molekuly **H_2O** $\Gamma_{red} = 3\,A_1 + 1\,A_2 + 3\,B_1 + 2\,B_2$ (9 stupňů volnosti)

transformace 3 atomů na každém 3 kartézské souřadnice

rotace $A_2 + B_1 + B_2$

translace $A_1 + B_1 + B_2$

vibrace $2\,A_1 + B_1$ - aktivní jak v IČ, tak Raman

Grupa C_{2v}

C_{2v}	E	C_2	$\sigma_v(xz)$	$\sigma_v(yz)$	_	_
A_1	1	1	1	1	z	$x^2; y^2; z^2$
A_2	1	1	-1	-1	R_z	xy
B_1	1	-1	1	-1	$x; R_y$	xz
B_2	1	-1	-1	1	$y; R_x$	yz

řád grupy 4

charaktery v reduc.

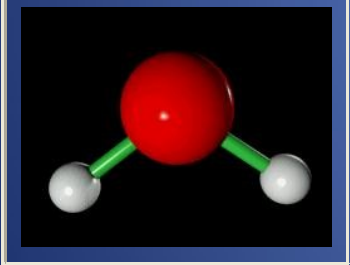
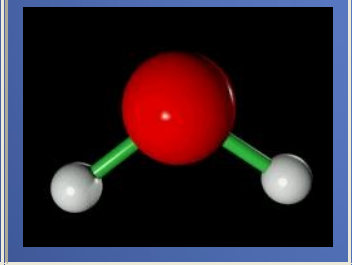
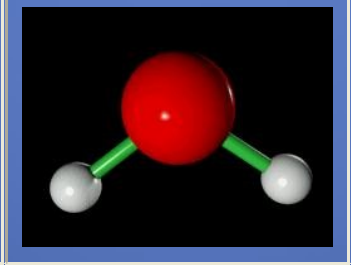
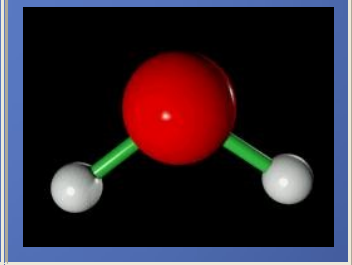
←charaktery v ired.

$9 - 1 + 3 + 1 = 12 \rightarrow 3\ A_1$

$9 - 1 - 3 - 1 = 4 \rightarrow 1\ A_2$

$9 + 1 + 3 - 1 = 12 \rightarrow 3\ B_1$

$9 + 1 - 3 + 1 = 8 \rightarrow 2\ B_2$

Identita	$C_2(z)$ rotace	σ (xz) zrcadlení	σ'(yz) zrcadlení
			
NUA = 3	1	3	1
CSO = 3	-1	1	1
Γ (NUA*CSO) = 9	-1	3	1

Grupa C_{2v}

C_{2v}	E	C_2	$\sigma_v(xz)$	$\sigma_v(yz)$	_	_
A_1	1	1	1	1	z	x^2; y^2; z^2
A_2	1	1	-1	-1	R_z	xy
B_1	1	-1	1	-1	x; R_y	xz
B_2	1	-1	-1	1	y; R_x	yz

– příklad - **CH_2Cl_2 Γ_{red} = 5 A_1 ✚ 2 A_2 ✚ 4 B_1 ✚ 4 B_2** (15 stupňů volnosti)

rotace **A_2 ✚ B_1 ✚ B_2**

translace **A_1 ✚ B_1 ✚ B_2**

vibrace **4 A_1 ✚ A_2 ✚ 2 B_1 ✚ 2 B_2** (9 stupňů volnosti)

4 **A_1** - aktivní jak v IČ, tak Raman

1 **A_2** - aktivní Raman

2 **B_1** - aktivní jak v IČ, tak Raman

2 **B_2** - aktivní jak v IČ, tak Raman

Grupa $\mathcal{D}_{4h}$

D_{4h}	E	$2C_4$	C_2	$2C'_2$	$2C''_2$	i	$2S_4$	σ_h	$2\sigma_v$	$2\sigma_d$		
A_{1g}	1	1	1	1	1	1	1	1	1	1		$x^2+y^2;\ z^2$
A_{2g}	1	1	1	-1	-1	1	1	1	-1	-1	R_z	
B_{1g}	1	-1	1	1	-1	1	-1	1	1	-1		x^2-y^2
B_{2g}	1	-1	1	-1	1	1	-1	1	-1	1		xy
E_g	2	0	-2	0	0	2	0	-2	0	0	$(R_x;R_y)$	$(xz;yz)$
A_{1u}	1	1	1	1	1	-1	-1	-1	-1	-1		
A_{2u}	1	1	1	-1	-1	-1	-1	-1	1	1	z	
B_{1u}	1	-1	1	1	-1	-1	1	-1	-1	1		
B_{2u}	1	-1	1	-1	1	-1	1	-1	1	-1		
E_u	2	0	-2	0	0	-2	0	2	0	0	$(x;y)$	

TEORETICKÉ VÝPOČTY VIBRAČNÍCH MODŮ

- **KVANTOVĚ CHEMICKÉ VÝPOČTY**
  - ***ab initio***
  - ***empirické***
    * ROVNOVÁŽNÉ POLOHY ATOMŮ
    * HMOTNOST ATOMŮ
    * SILOVÉ POLE MOLEKULY (SÍLA VAZEB)

- **APROXIMACE PŘI VÝPOČTECH**
  - VLIV RŮZNÝCH TYPŮ INTRA- A INTER-MOLEKULÁRNÍCH INTERAKCÍ

TruScan

Technology Overview/Introduction

Smaller Systems

Delta Nu/Smiths

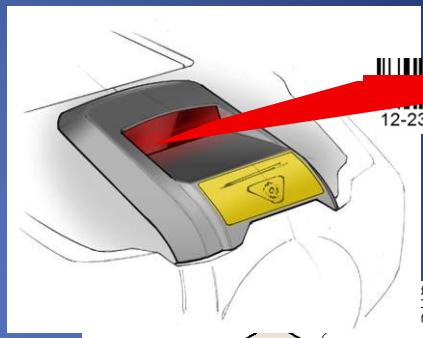
OceanOptics

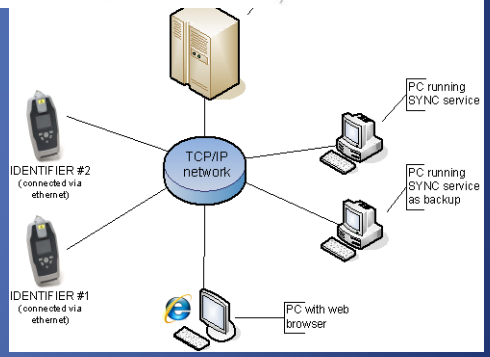
BWTek

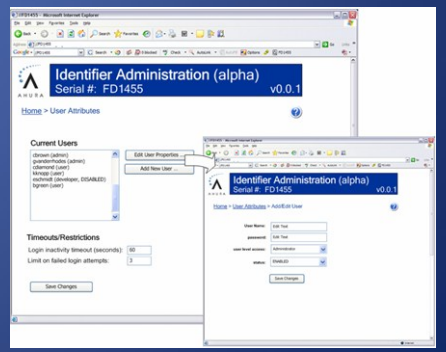
InPhotonics

TruScan

- **Technology:** Raman spectroscopy + embedded analytics
- **Designed for Solid & Liquid Authentication and Discovery**
  - Site specific material library
- **Battery Charge Lifetime**
  - > 5 hrs continuous operation
- **Rugged & Lightweight**
  - Water-resistant & <4 lb
  - -20 °C to +40°C Operation
  - Mean Time between failure MTBF >6 years
- **Conducive to 21CFR11 compliance**
  - User-level access control
  - Strictly Managed workflow
  - E-records contain all pertinent information
- **IQ/OQ documentation**
- **Integrated Barcode scanner**
  - Directly capture barcode information from material and/or batch/container
- **Fleet management/Web-based administration**
  - no PC client to learn or manage; manage a system from any PC with a web-browser
  - Deploy group of methods to any unit in the field
- **Ethernet Sync via TCP/IP**
  - automated data delivery & report generation to a network location of choice
- **User built material library (>4000 compounds in discovery library)**







point-and-shoot

barcode reader

Barcode standards (MOST COMMON)

LISTED IN APPENDIX F OF USER MANUAL

Pharmaceutical Industry Drivers

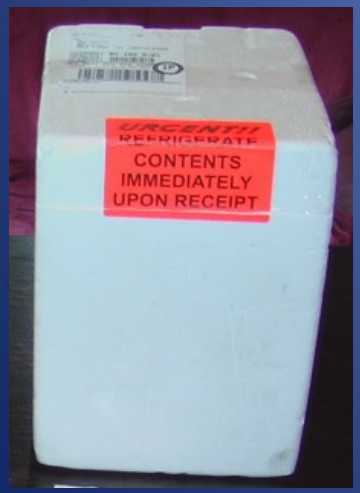
- **Tightening Regulatory Control for Raw Materials**
  - Good Manufacturing Practices (GMPs) applied to Active Pharmaceutical Ingredients (API) production
  - European Medicine Agency (EMEA) 100% inspection requirement
  - Food and Drug Administration (FDA) desire to move toward 100% inspection

- **Lean Manufacturing and Just-In-Time**
  - Profits must be generated from manufacturing efficiency gains
  - Increasing competition (Low Cost Regions (LCRs) and Contract Manufacturing Organizations (CMOs))
  - Continuous pressure to speed results and use resources more efficiently (warehouse space, personnel, capacity, etc.)

- **Quality Initiatives**
  - Process Analytical Technology (PAT) initiative to identify and measure critical quality attributes
  - Move towards *Ensuring Quality* as opposed to testing for it
  - Six sigma, Quality by Design (QbD), etc

Sampling Process

open container → document → transfer → label → protect → close container → quarantine lot → ship →



The dreaded FDA-483 Warning letter

DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
Silver Spring, MD 20993

APR 2 1 2008

Warning Letter

Via FedEx and facsimile

Dr. Yan Wang, Ph.D.
General Manager
Changzhou SPL Company, Ltd (a/k/a "Kaipu")
3 Changhong West Road
Hutang Township, Wujin City
Changzhou
China

2. You fail to have adequate systems for evaluating the suppliers of heparin crude materials, and the crude materials themselves, to ensure that these materials are acceptable for use.

Our inspection found (Observation #6 of the FDA-483) that you received lots of material from an unacceptable workshop vendor that were used in your API. In your March 17, 2008, response to observation #6, your firm acknowledges inadequacies in the firm's supplier qualification efforts. For example, you state that the firm received and used heparin crude materials from a workshop that had been designated by your firm in a "pre-audit" as "unacceptable" and that was ultimately not approved by your firm. Your firm used this crude material in the production of API lots that were shipped to the United States.

Your system for evaluating suppliers of crude heparin material is ineffective to ensure that materials are acceptable for use. As described above, your firm accepted and used heparin crude material from a supplier that you had preliminarily determined was unacceptable. Your system failed to verify that the supplier was acceptable prior to the use of the crude material. Furthermore, after your firm determined that the supplier was

[redacted]

hat are received and used in producing heparin sodium API should be qualified using a system to ensure that raw materials are of acceptable identity, quality and purity before use. It is important to establish appropriate specifications for these materials and to assure your suppliers provide materials meeting these specifications. These specifications should be approved by the quality unit. Your firm has failed to establish appropriate specifications for your incoming crude materials.

Structured workflow streamlines ID testing

Start Shift:

System Suitability

During shift:

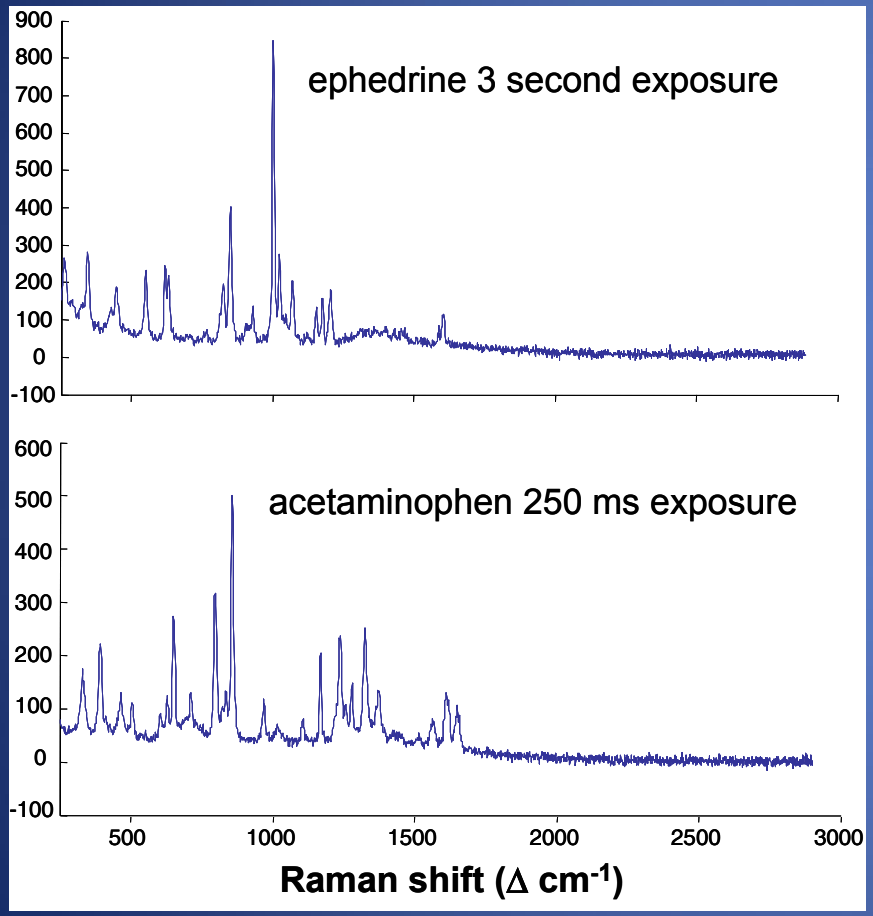
Select method →
Barcode sample →
Scan through bag →
View results

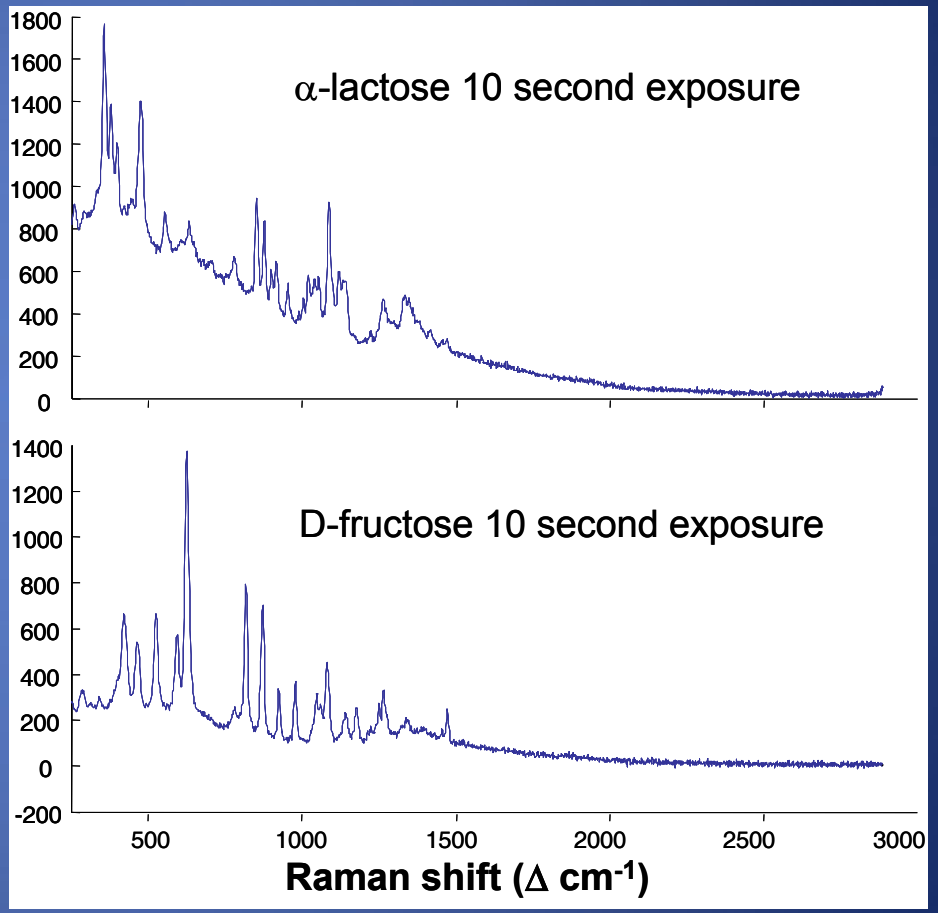
End Shift:

Sync to Archive

TruScan reduces sample cycle times to seconds or minutes

Speed of Sampling





TruScan brings analysis to the sample

Measure where Required

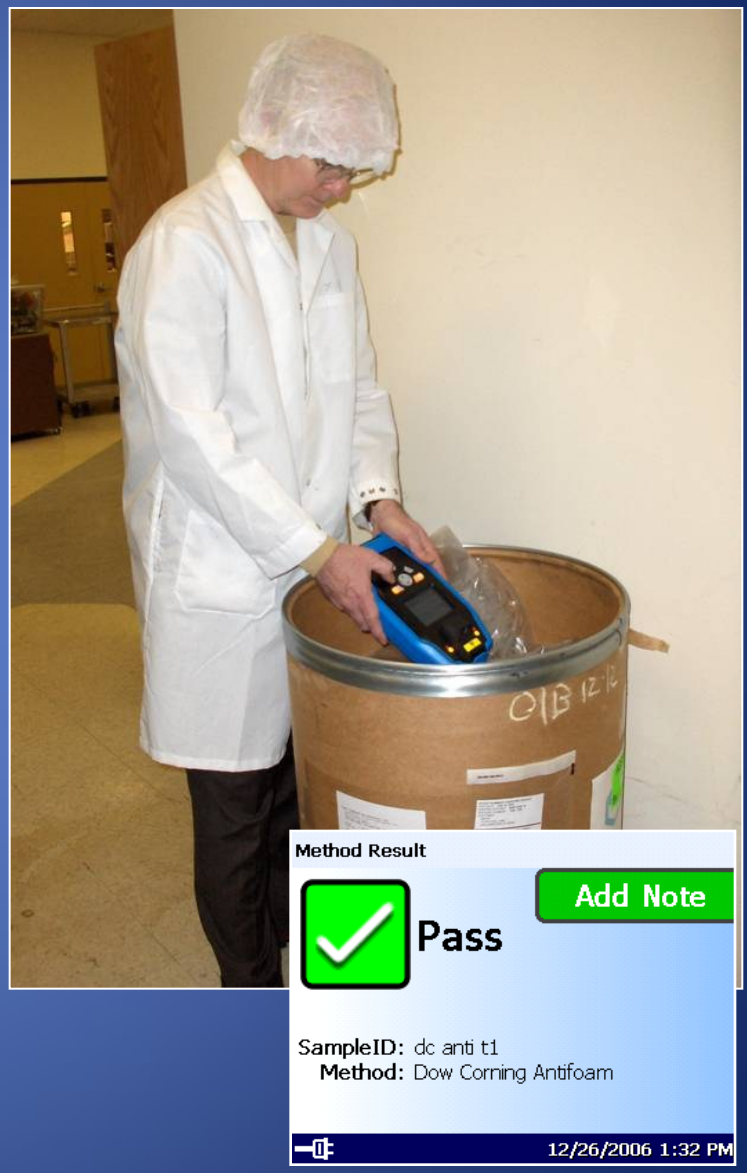
- Avoids need to take sample to lab
- Environment independence
- Self-contained
- Rugged and reliable

Measure in Container

- Avoids contamination
- Avoids exposure to potent materials
- Fast

Decision Capable Instrument

- Avoids extensive training requirements
- Reproducible, operator-independent results



What is TruScan for?:
Recent Literature on Antimalarial drugs

OPEN ACCESS Freely available online

PLoS MEDICINE

Policy Forum

Manslaughter by Fake Artesunate in Asia—Will Africa Be Next?

Paul N. Newton*, Rose McGready, Facundo Fernandez, Michael D. Green, Manuela Sunjio, Carinne Bruneton, Souly Phanouvong, Pascal Millet, Christopher J. M. Whitty, Ambrose O. Talisuna, Stephane Proux, Eva Maria Christophel, Grace Malenga, Pratap Singhasivanon, Kalifa Bojang, Harparkash Kaur, Kevin Palmer, Nicholas P. J. Day, Brian M. Greenwood, François Nosten, Nicholas J. White

Falciparum malaria kills, and it particularly kills the rural poor. Artemisinin derivatives, such as artesunate, are a vital component of *Plasmodium falciparum* malaria treatment and control in the face of globally increasing antimalarial drug resistance. Since 1998 a worsening epidemic of sophisticated counterfeit "artesunate" tablets (containing no artesunate) has plagued mainland Southeast Asia (see Figure S1). In some countries, most of the available artesunate is fake [1–5].

Artemisinin derivatives are remarkably rapid in their antimalarial effects, and they are very well tolerated. So where these medicines are available, they are sought after. But as they are relatively expensive, a demand is created for cheaper versions amongst the poorest and most vulnerable people, upon whom the counterfeiters have preyed–with fatal results.

Documented Death due to Fake Artesunate

The death of patients with untreated falciparum malaria, as a result of unwittingly taking fake artesunate, is hidden in the inadequately documented mortality statistics of the relatively voiceless rural poor. But there is no doubt that such deaths occur, and they are probably common.

In February 2005, a 28-year-old man presented with fever to a rural hospital in eastern Burma where he was diagnosed as having uncomplicated hyperparasitaemic falciparum malaria by microscopy (4.2% infected red blood cells). He was treated with oral artesunate, labelled as made by Guilin Pharmaceutical (Guangxi, People's Republic of China), 4 mg/kg once a day, the treatment of choice in this area. Since artemisinin derivatives have been used in this area, not one of 600 patients prospectively studied with ≥4% parasitaemia has died [6]. However, on the third night the young Burmese man became unconscious and was transferred to another hospital where he was found to be in a coma

DOI: 10.1371/journal.pmed.0030197.g001

Figure 1. Genuine (Left) and Counterfeit (Right) Cotexcin (Dihydroartemisinin) from Tanzania (Photograph by Manuela Sunjio)

The Policy Forum allows health policy makers around the world to discuss challenges and opportunities for improving health care in their societies.

Funding: P. Newton, R. McGready, S. Proux, N. Day, F. Nosten, and N. White are supported by the Wellcome Trust of Great Britain. C. Whitty, H. Kaur, and B. Greenwood are supported by the Bill and Melinda Gates Foundation through the Gates Malaria Partnership. The authors received no specific funding to write this article.

Competing Interests: The authors declare that they have no competing interests. The authors' funding bodies had no role in the preparation of this article.

Citation: Newton PN, McGready R, Fernandez F, Green MD, Sunjio M, et al. (2006) Manslaughter by fake artesunate in Asia—Will Africa be next? PLoS Med 3(6): e197. DOI: 10.1371/journal.pmed.0030197

DOI: 10.1371/journal.pmed.0030197

Abbreviations: ACT, artemisinin derivative–based combination therapy

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PLoS Medicine **3**:752-755 (2006)

JOURNAL OF RAMAN SPECTROSCOPY
J. Raman Spectrosc. 2007; 38: 181–187
Published online 18 October 2006 in Wiley InterScience
(www.interscience.wiley.com) DOI: 10.1002/jrs.1621

JRS

Fast detection and identification of counterfeit antimalarial tablets by Raman spectroscopy

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Received 7 January 2006; Accepted 21 April 2006

During the last decade there has been an apparent increase in the prevalence of counterfeit medicines in developing as well as developed countries. The pivotal antimalarial artesunate has been counterfeited on a large scale in SE Asia. In this work, the possibilities of Raman spectroscopy are explored as a fast and reliable screening method for the detection of counterfeit artesunate tablets. In this study, 50 'artesunate tablets', purchased in SE Asia, were examined. This spectroscopic method was able to distinguish between genuine and counterfeit artesunate and to identify the composition of the counterfeit tablets. These contained no detectable levels of artesunate, but consisted mostly of starch, calcite ($CaCO_3$), and paracetamol (4-acetamidophenol). In one particular case an admixture of rutile (TiO_2) and artesunate was detected. The results of the investigation by Raman spectroscopy were in agreement with those of colorimetric tests and of liquid chromatography-mass spectrometry on the artesunate. Moreover, principal components analysis (PCA) was combined with hierarchical cluster analysis to establish an automated approach for the discrimination between different groups of counterfeits and genuine artesunate tablets. These results demonstrate that Raman spectroscopy combined with multivariate analysis is a promising and reliable methodology for the fast characterization of genuine and counterfeit artesunate antimalarial tablets. Copyright © 2006 John Wiley & Sons, Ltd.

KEYWORDS: artesunate; counterfeit drug detection; malaria: principal components analysis (PCA)

INTRODUCTION

Counterfeit medicines happen to be a problem not only for many developing nations but for developed countries as well.[1] The United States Food and Drug Administration (FDA) estimates that counterfeits make up to more than 10% of the global medicines market.[2] In 2004, FDA's Office of Criminal Investigations initiated 58 counterfeit drug cases in the United States compared to nine cases in 1997, which shows that counterfeit drugs are an increasing problem.[3]

Counterfeit medicines include drugs without sufficient active ingredient, without any active ingredients, or with fake packaging.[1,2] In wealthier countries new and expensive medicines, such as hormones, corticosteroids, and cancer drugs, are frequently counterfeited.[4] Among the most well-known counterfeit drugs in wealthier countries are Viagra and Cialis, which are used against erectile dysfunction.[1,5] In developing countries, the most counterfeited medicines are those used to treat widespread infectious diseases such as tuberculosis and malaria.[4]

Until recently, analytical controls in the drug distribution system were considered to be supplementary[6] to the quality control at the production unit. Counterfeit drug detection is often a complex process. Careful visual inspection of the product's packaging and labeling is the first step of defence, since modifications may indicate a potential counterfeit. Visual inspection is followed by chemical analysis, often

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Journal of Raman Spectroscopy **18**:181-187 (2007)

Example Artesunate Tablet Report

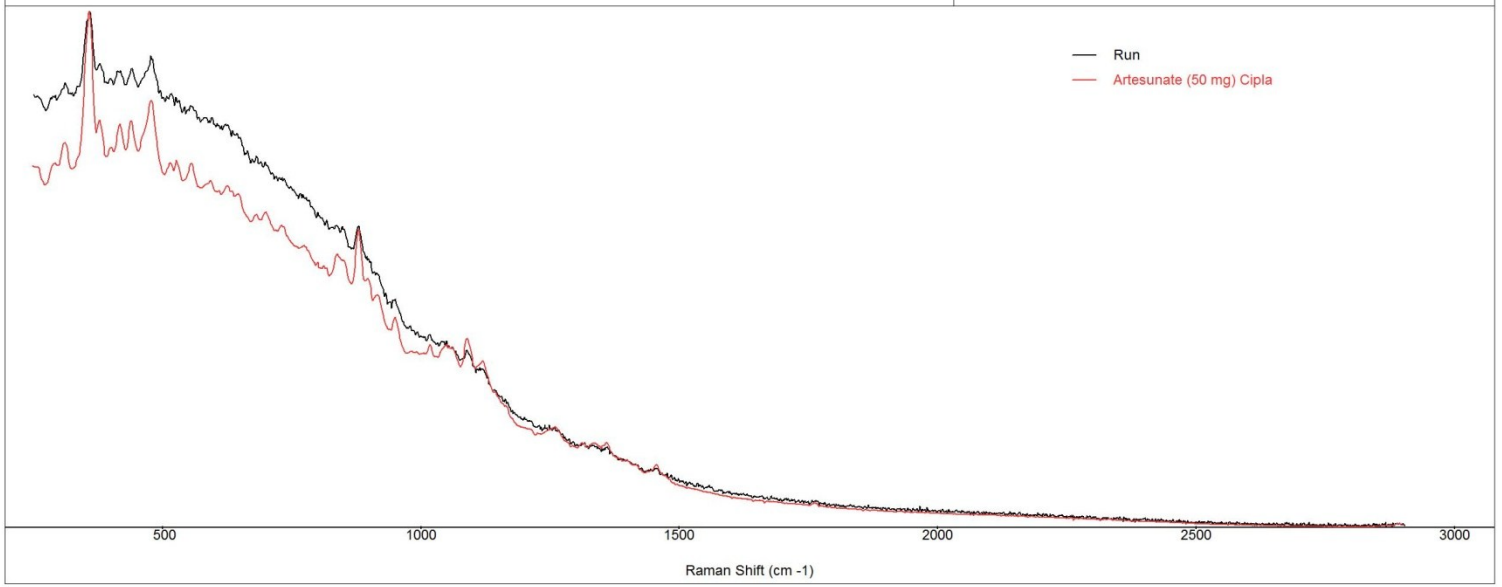
Run Label:	Run-Robert C. Brush-20080422-171613
Run Time:	2008-04-22 17:16:13
Device Serial:	TS1059
Method:	Artesunate (50 mg) Cipla
Sample ID:	t5a
Pval:	0.168380
Note:	
Result:	Pass

AHURA

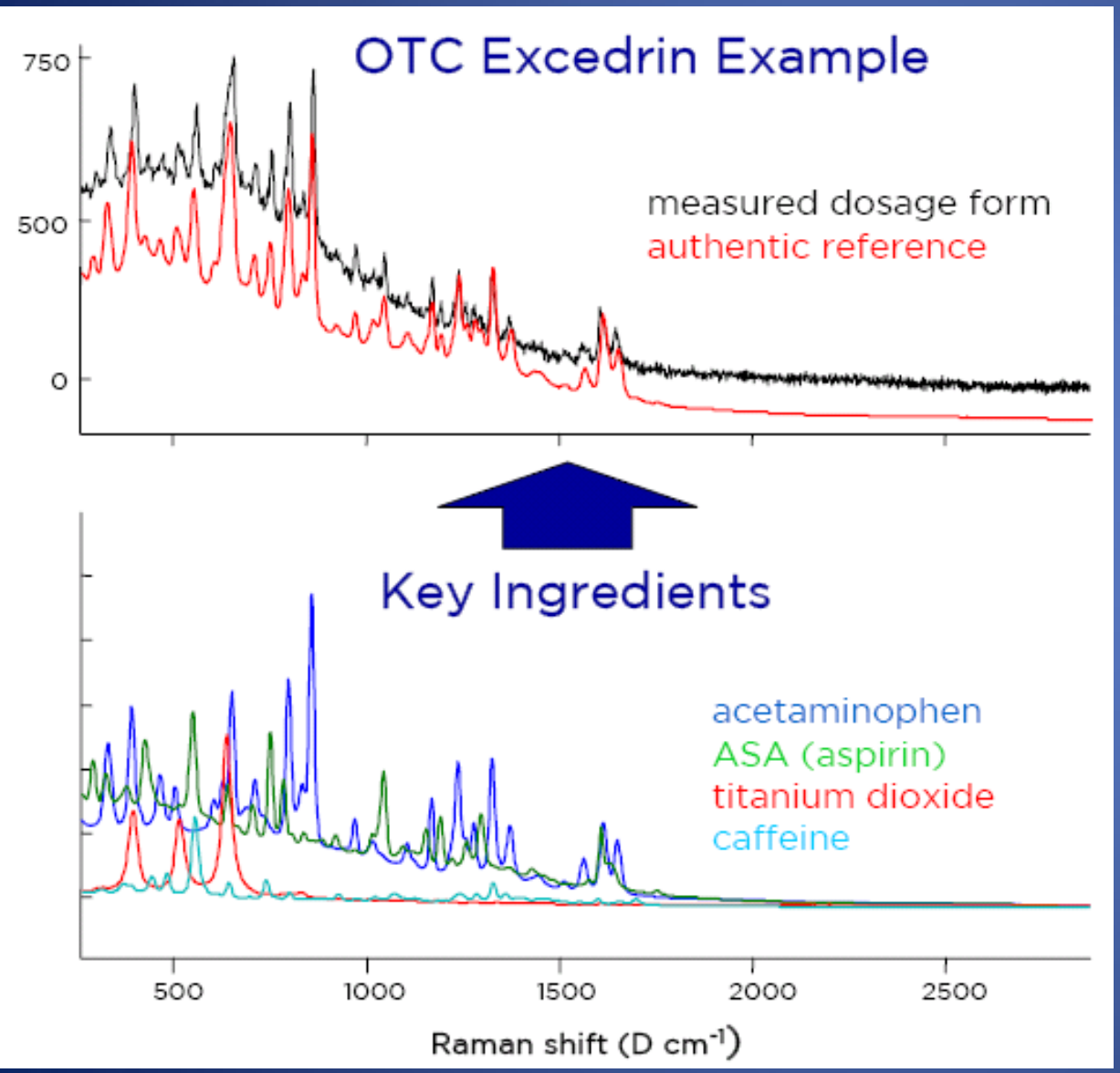
AHURA
TruScan Report

User Name:	Robert C. Brush
Login Time:	2008-04-22 10:32:19
Software Version:	1.1.1 (Build 4444)
Last Tested:	2008-04-22 08:56:18
Last Test Result:	Pass
Calibration Check:	Pass
CCD Check:	Pass
Laser Power:	Pass

Warnings: none



TruScan creates unique spectral signature with all elements of a formulation



Effective

TruScan measures the relative values of:

API, excipients, coatings, dyes, and even impurities

The combination and relative value of these ingredients provides a unique spectral ID

Secure

Spectral ID lacks sufficient detail to be effective for reverse engineering

Where to use TruScan?

Increasing Utility

Customs
- Partnership to provide Customs with proprietary methods
- No need to go on-site
- Saves time & money

Forensic Labs
- Expedite processing of samples received → increase throughput, reduce delays
- Used adjunctively with other technologies

Manufacturer

Field Investigations (reactive)
- Respond on-site to suspicious pharmaceuticals
- Allows instant, remote determination of counterfeits
- Brings analytical capabilities into suspect facility
- Allows forensic research and tracking of counterfeits
- Facilitate quicker identification and arrest

Pre-Shipment Inspection
- Partnership with Customs to expedite processing
- Partnership with 3rd party inspection services (SGS, Cotecna, etc.)

Field Monitoring (proactive)
- Perform random inspections of pharmacies, markets, manufacturing facilities, and hospitals
- Allows instant remote determination of counterfeits
- Brings analytical capabilities into ANY facility
- Allows forensic research and tracking of counterfeits
- Facilitate quicker identification and arrest
- Acts as deterrent to counterfeiters

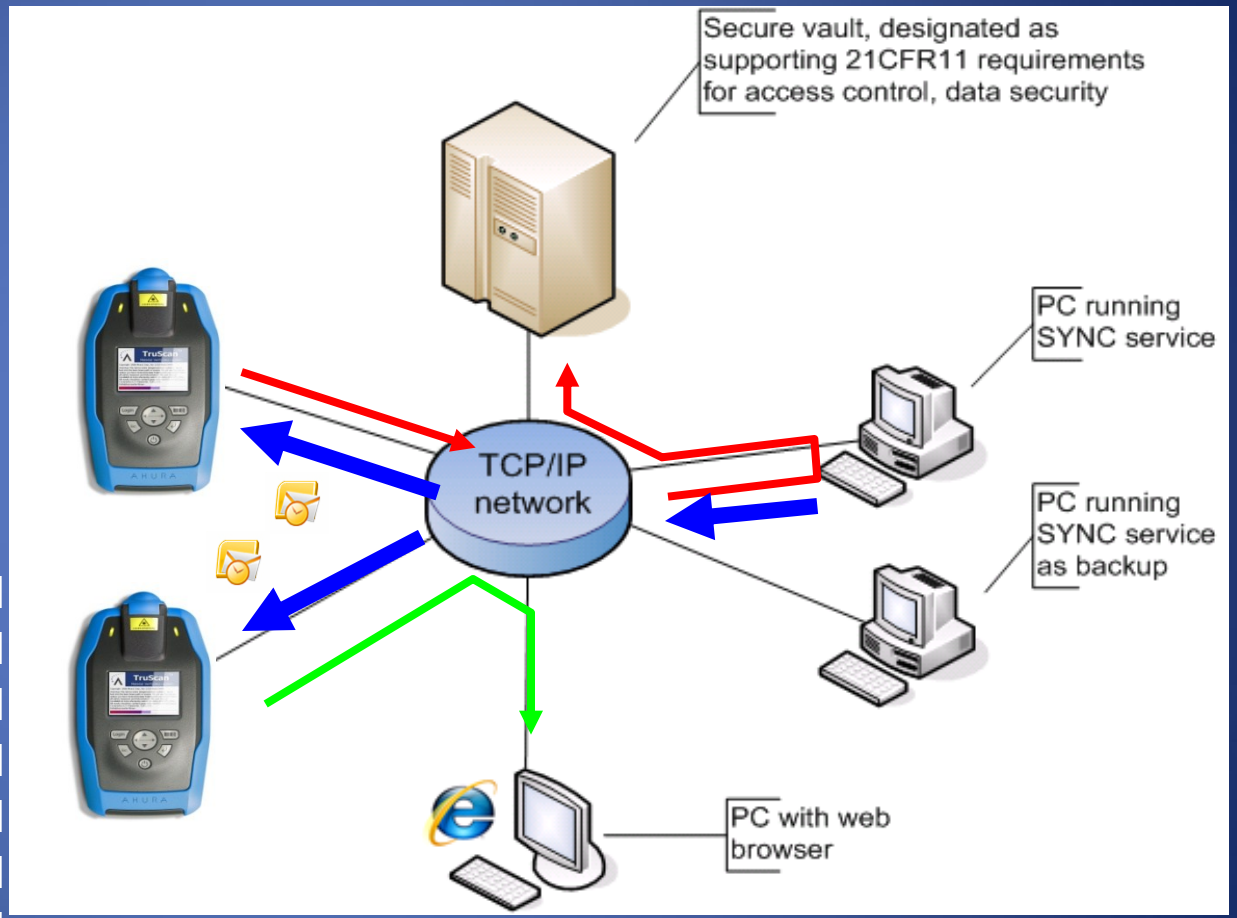
Supply Chain Inspection
- Partnership with Supply Chain Partners – wholesalers, distributors, etc.

Courtesy of Julien Bradley Ahura Scientific/ presentation at IMPACT 2008-WHO

System Neighborhood

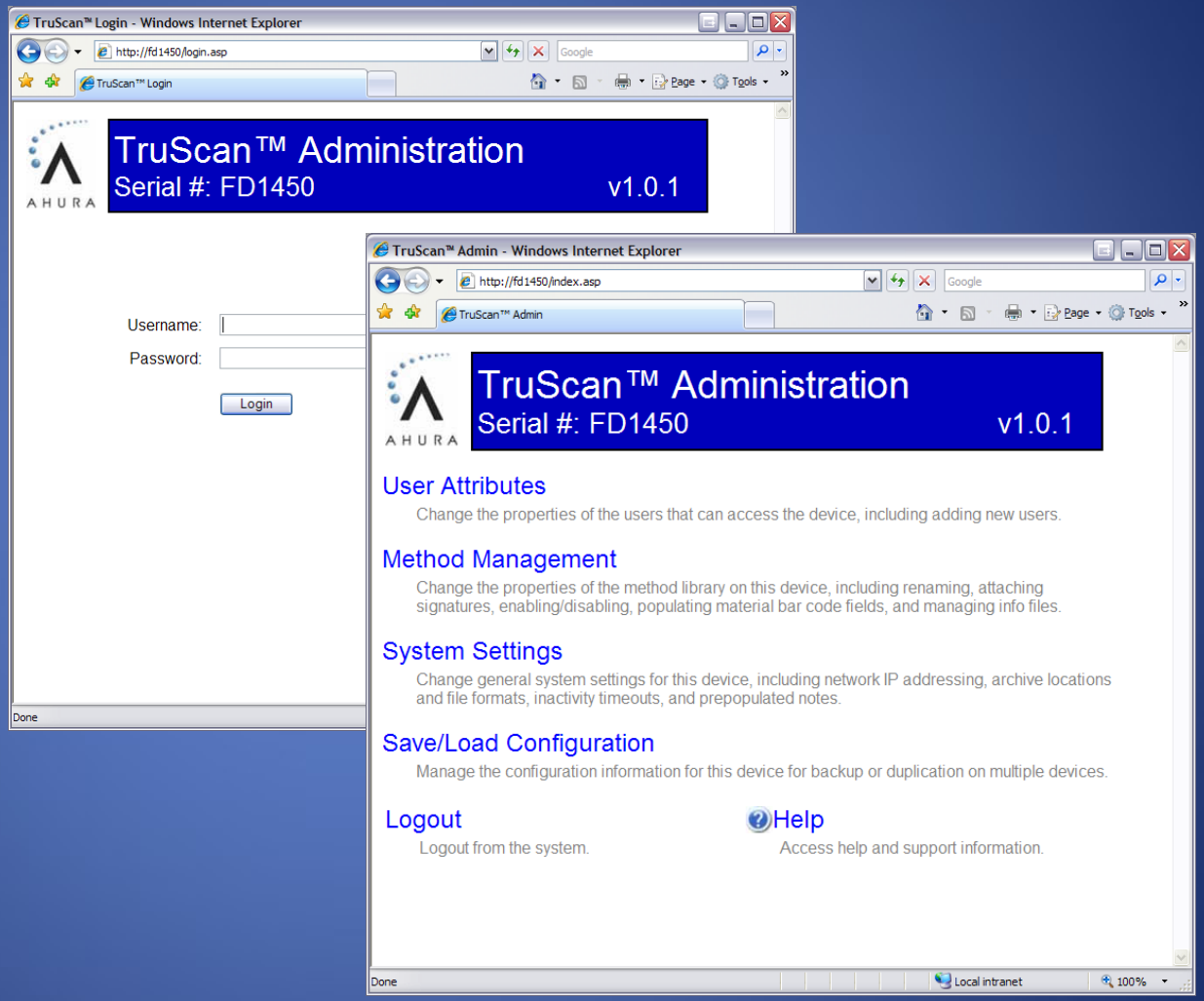
Central Administration through Corporate LAN

- System administration
- Method creation and control
- Method dissemination (Pack or individual)
- Central document repository
- Users credentials
- Remote Device configuration
- Report generation
- Traceability



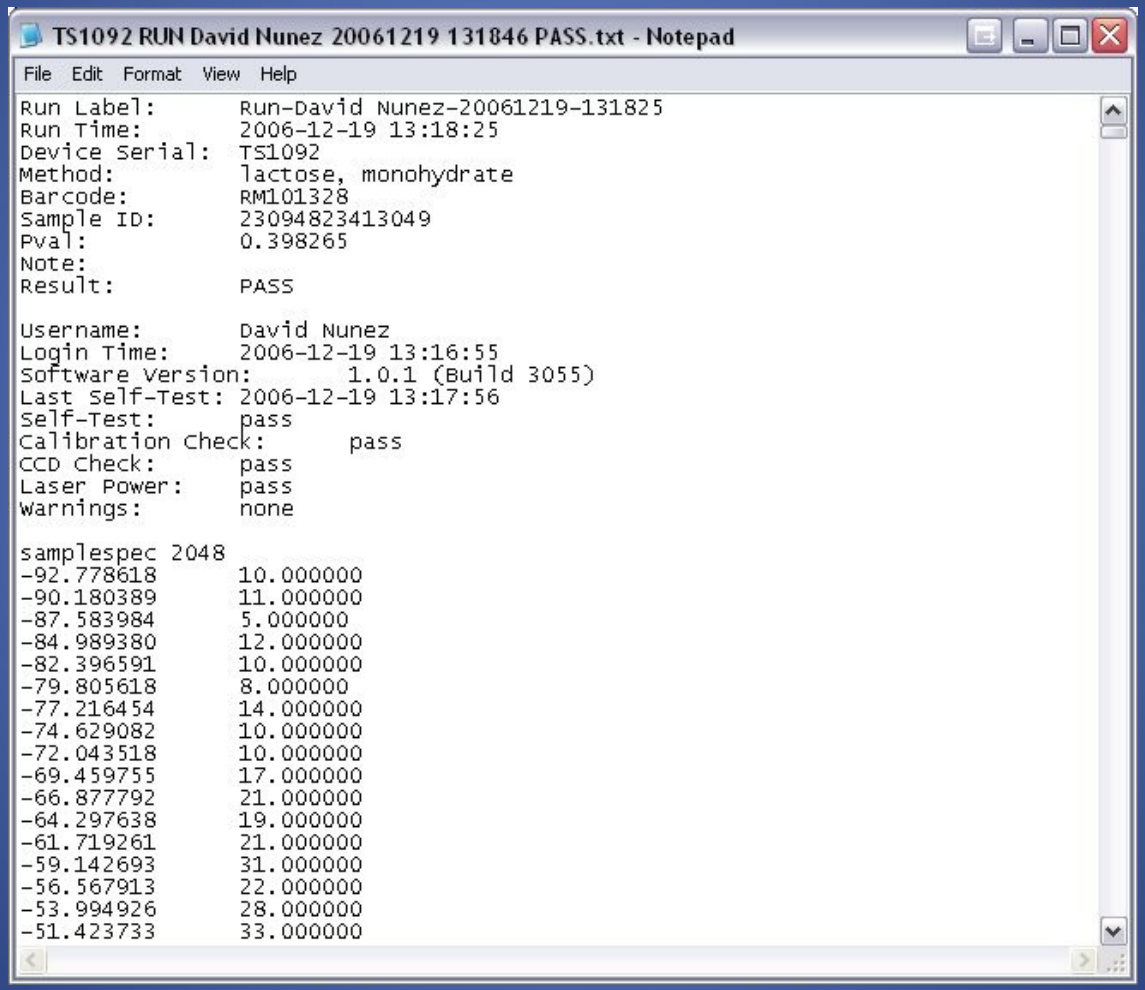
System Administration - WebAdmin

web browser
(no external net connection required)



Electronic Records – text

LIMS and e-notebook friendly …



TS1092 RUN David Nunez 20061219 131846 PASS.txt - Notepad

File Edit Format View Help

```
Run Label:      Run-David Nunez-20061219-131825
Run Time:       2006-12-19 13:18:25
Device Serial:  TS1092
Method:         lactose, monohydrate
Barcode:        RM101328
Sample ID:      23094823413049
Pval:           0.398265
Note:
Result:         PASS

Username:       David Nunez
Login Time:     2006-12-19 13:16:55
Software Version:       1.0.1 (Build 3055)
Last Self-Test: 2006-12-19 13:17:56
Self-Test:      pass
Calibration Check:      pass
CCD Check:      pass
Laser Power:    pass
Warnings:       none

samplespec 2048
-92.778618      10.000000
-90.180389      11.000000
-87.583984      5.000000
-84.989380      12.000000
-82.396591      10.000000
-79.805618      8.000000
-77.216454      14.000000
-74.629082      10.000000
-72.043518      10.000000
-69.459755      17.000000
-66.877792      21.000000
-64.297638      19.000000
-61.719261      21.000000
-59.142693      31.000000
-56.567913      22.000000
-53.994926      28.000000
-51.423733      33.000000
```

Electronic Records – PDF/JPEG

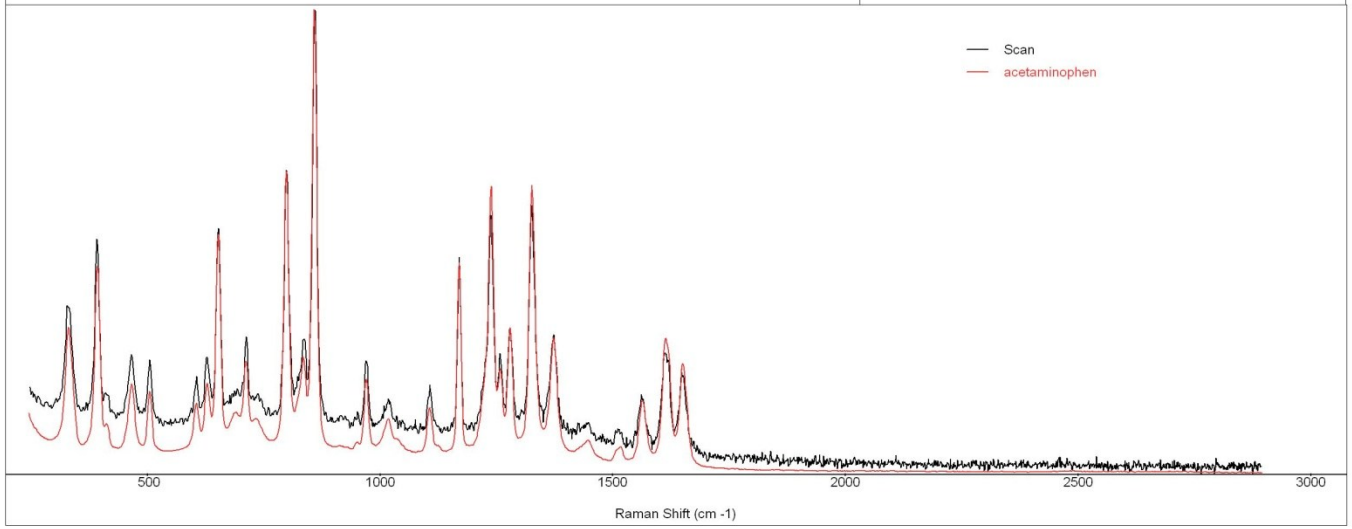
Run Label:	Run-Administrator-20061218-171602
Run Time:	2006-12-18 17:16:02
Device Serial:	TS1000
Method:	acetaminophen
Sample ID:	a0235
Pval:	0.356498
Note:	
Result:	PASS

AHURA

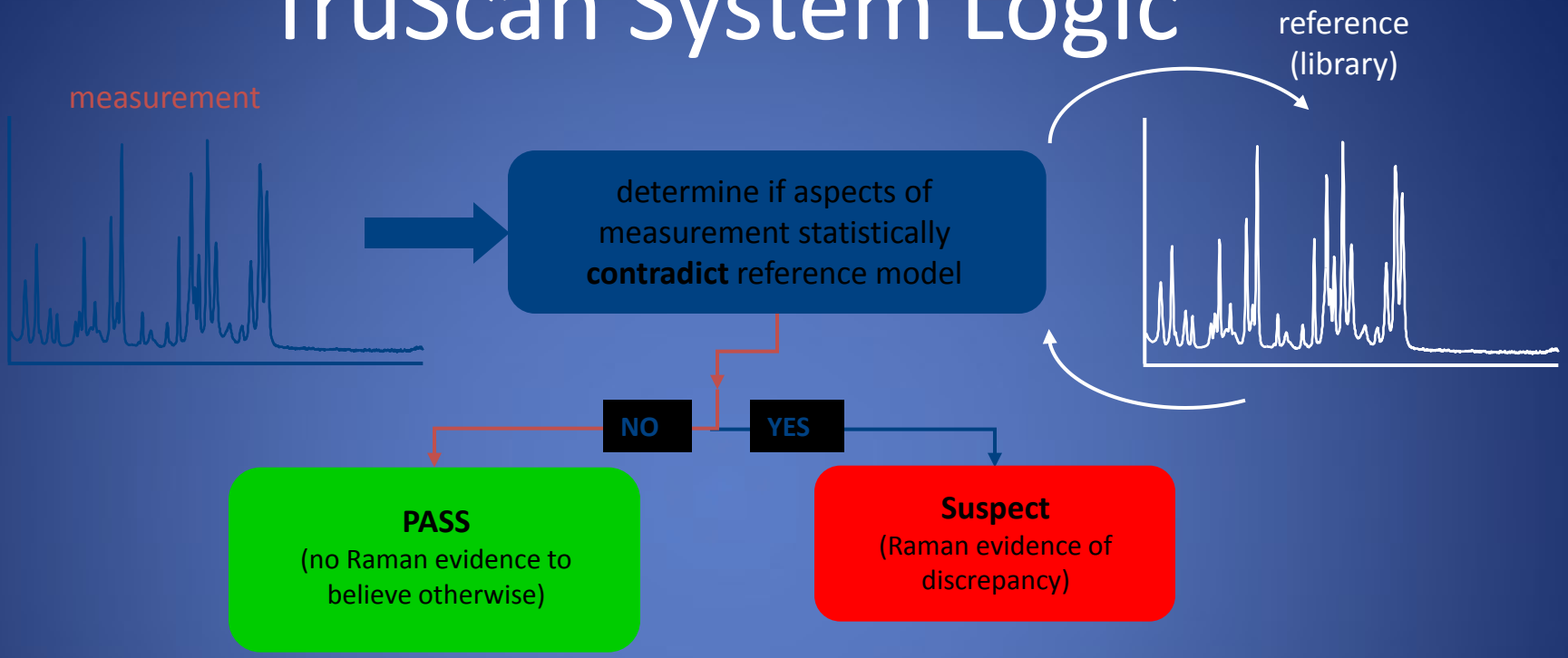
AHURA
TruScan Report

User Name:	Administrator
Login Time:	2006-12-18 17:14:12
Software Version:	1.0.0 (Build 2965)
Last Self-Test:	2006-12-18 17:15:13
Self-Test:	Pass
Calibration Check:	Pass
CCD Check:	Pass
Laser Power:	Pass

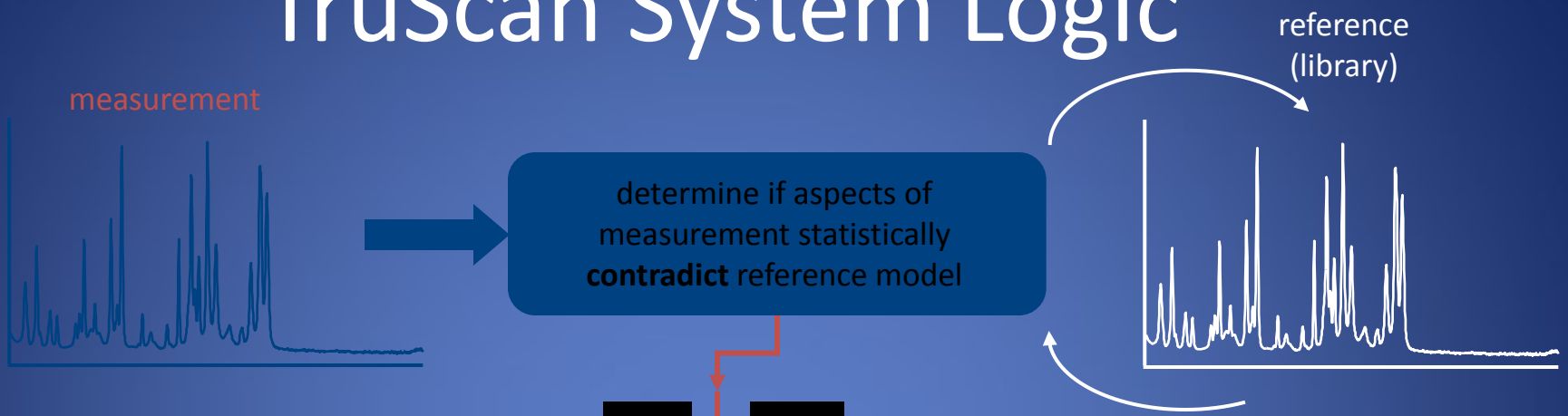
Warnings: none

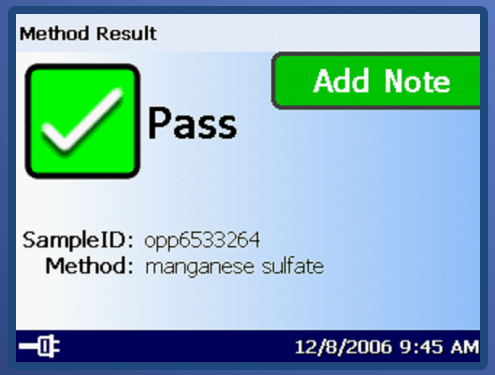


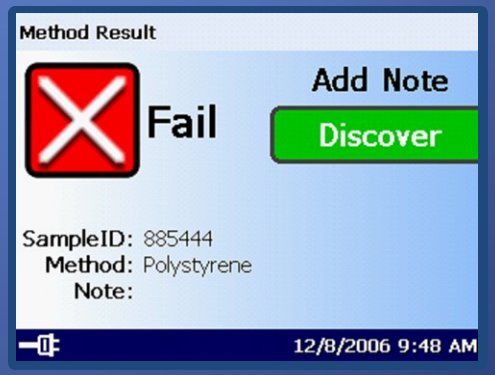
TruScan System Logic



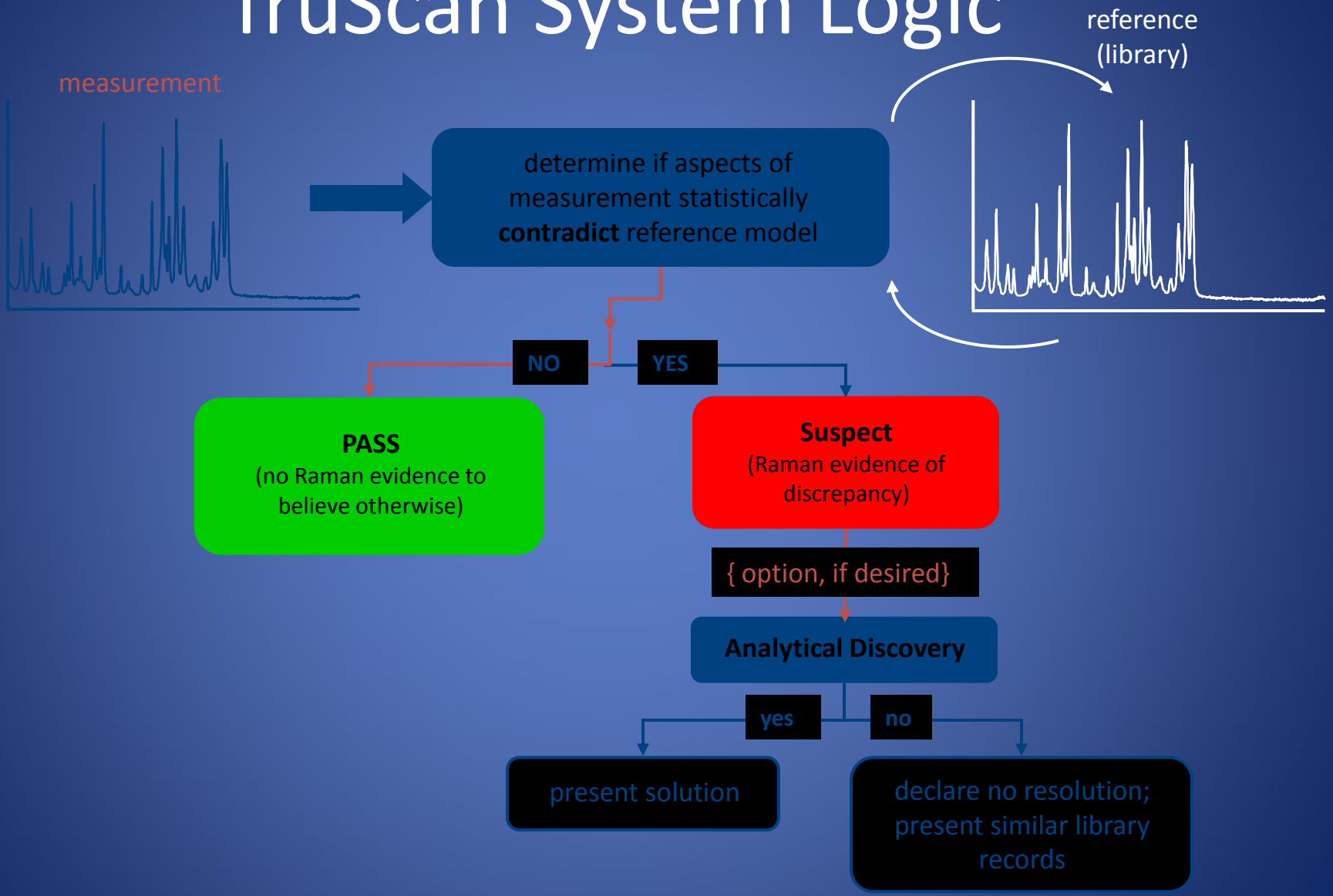
TruScan System Logic







TruScan System Logic

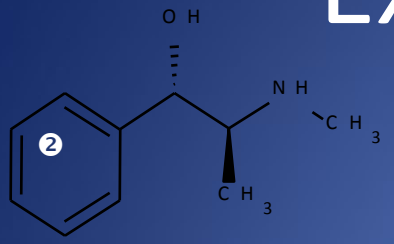


TruScan automates probabilistic analysis

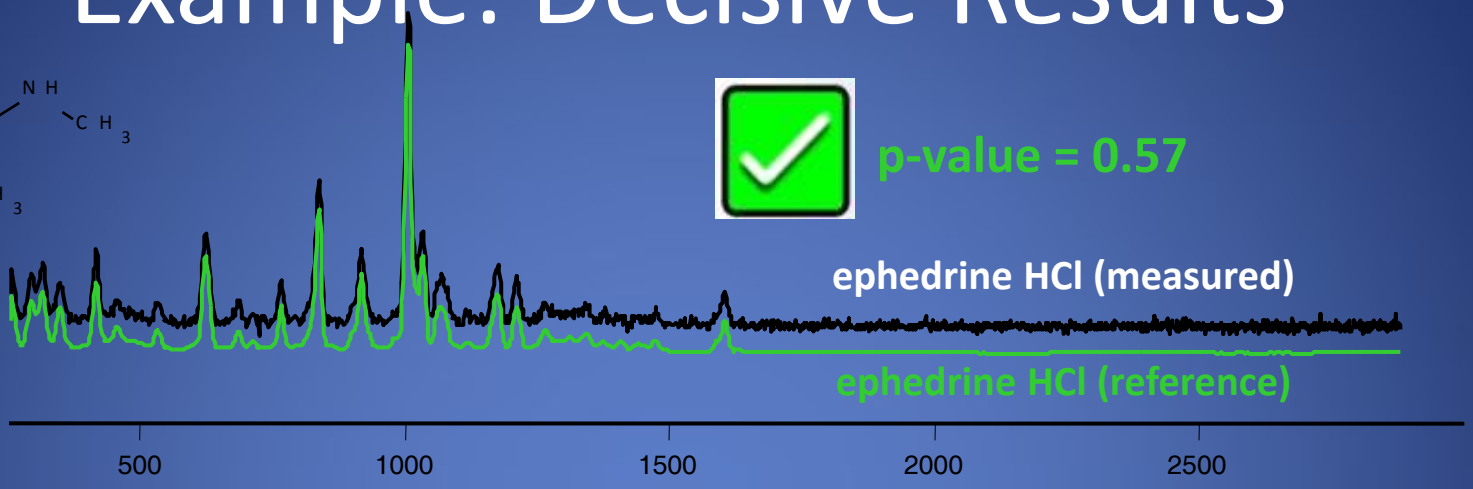
- p-value expresses degree of consistency between test material and reference (Equivalence of a T-Test with multiple variables)
- TruScan uses a p-value cutoff of 0.05 (95% confidence limit) to make decisions:

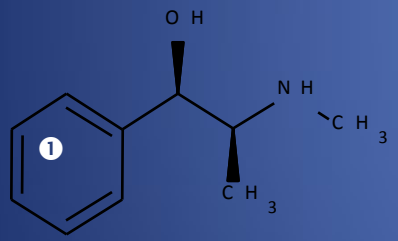
p value	Result
p< 0.05	FAIL (not a Match)
p> = 0.05	PASS (does Match)

Example: Decisive Results

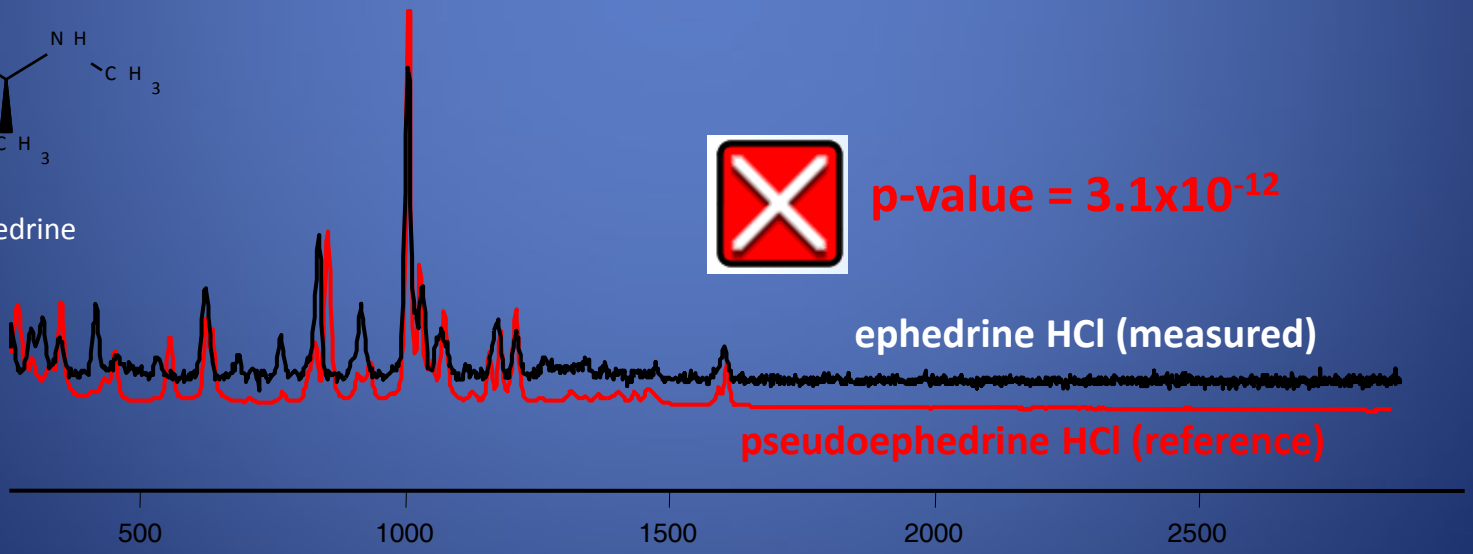


❷ ephedrine



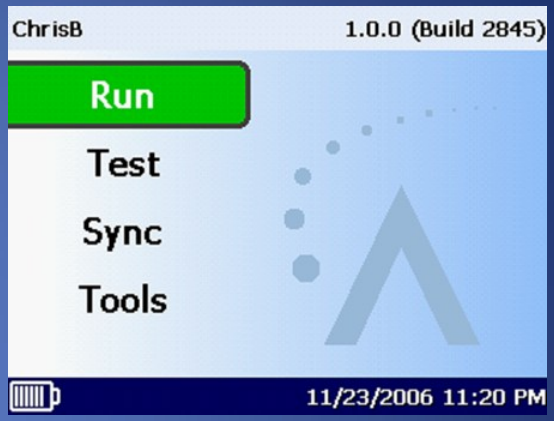


❶ pseudoephedrine



Validation and Regulatory Compliance

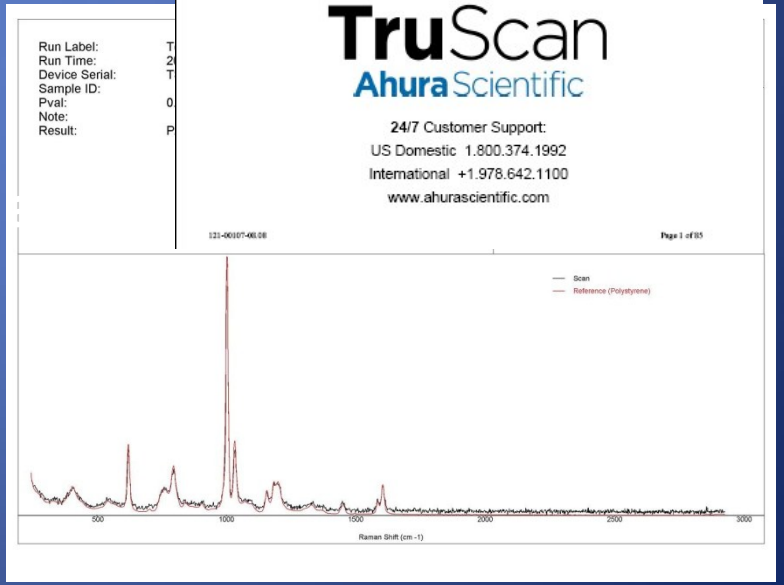
- Designed for use in 21 CFR 11 compliant environments
- Multilevel access control by username, password
  - 3 user levels: user, method developer, administrator
- Electronic records created for every test



- Records directly delivered to configurable vault destination on SYNC
- SYNC benefits from TCP/IP fault protection (ensures against packet-loss or corruption during transfer/archive) and transaction verification
- E-records, Audit Trail, Sync Summary directly delivered to configurable vault destination on SYNC

Instrument Qualification

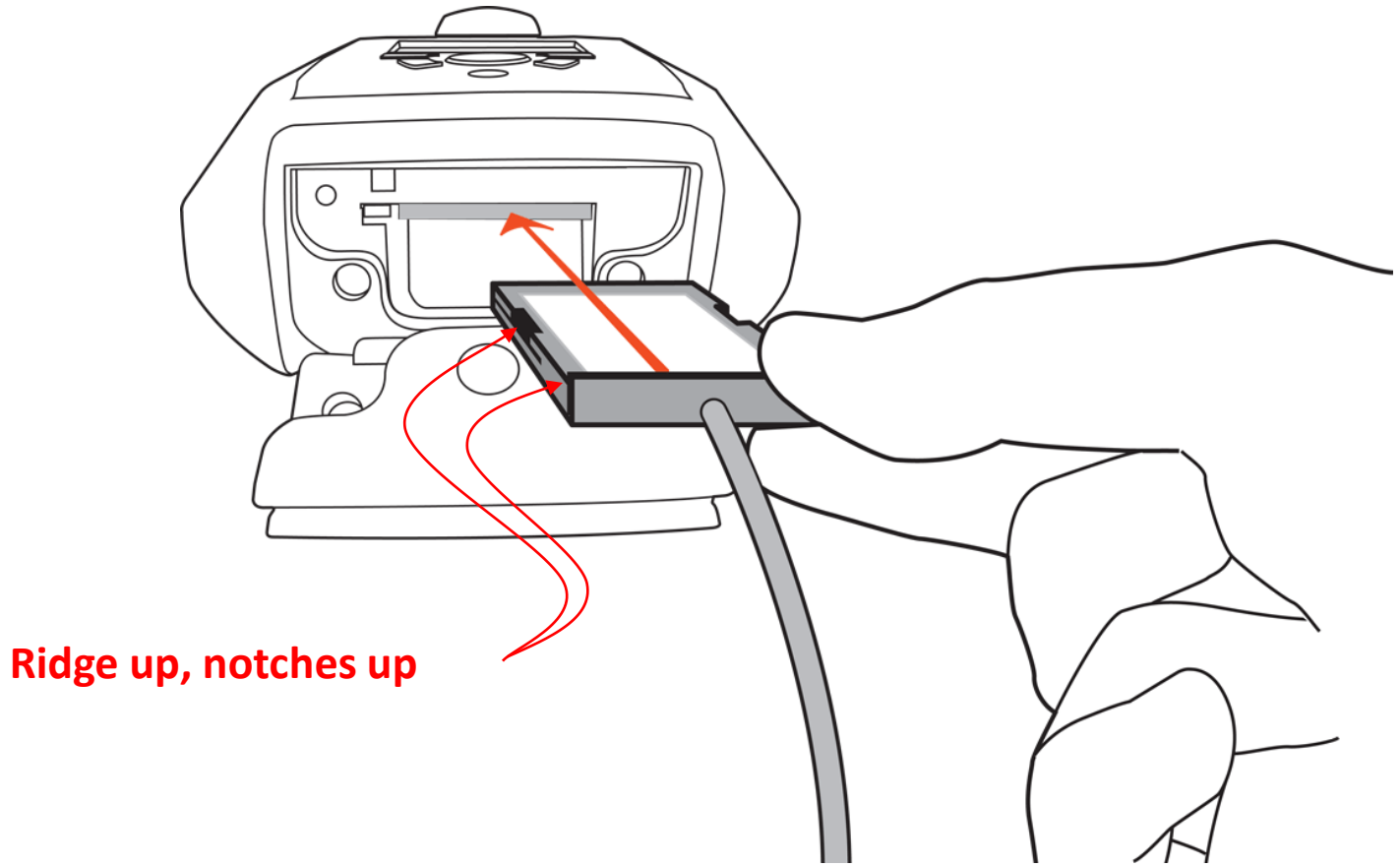
- Available Instrument Qualification- Operation Qualification (IQ/OQ) documentation and services
- Calibration and Performance Qualification (PQ) based on ASTM 1840 and USP <1120>
- Polystyrene Rod is system suitability standard
- PQ test takes few seconds



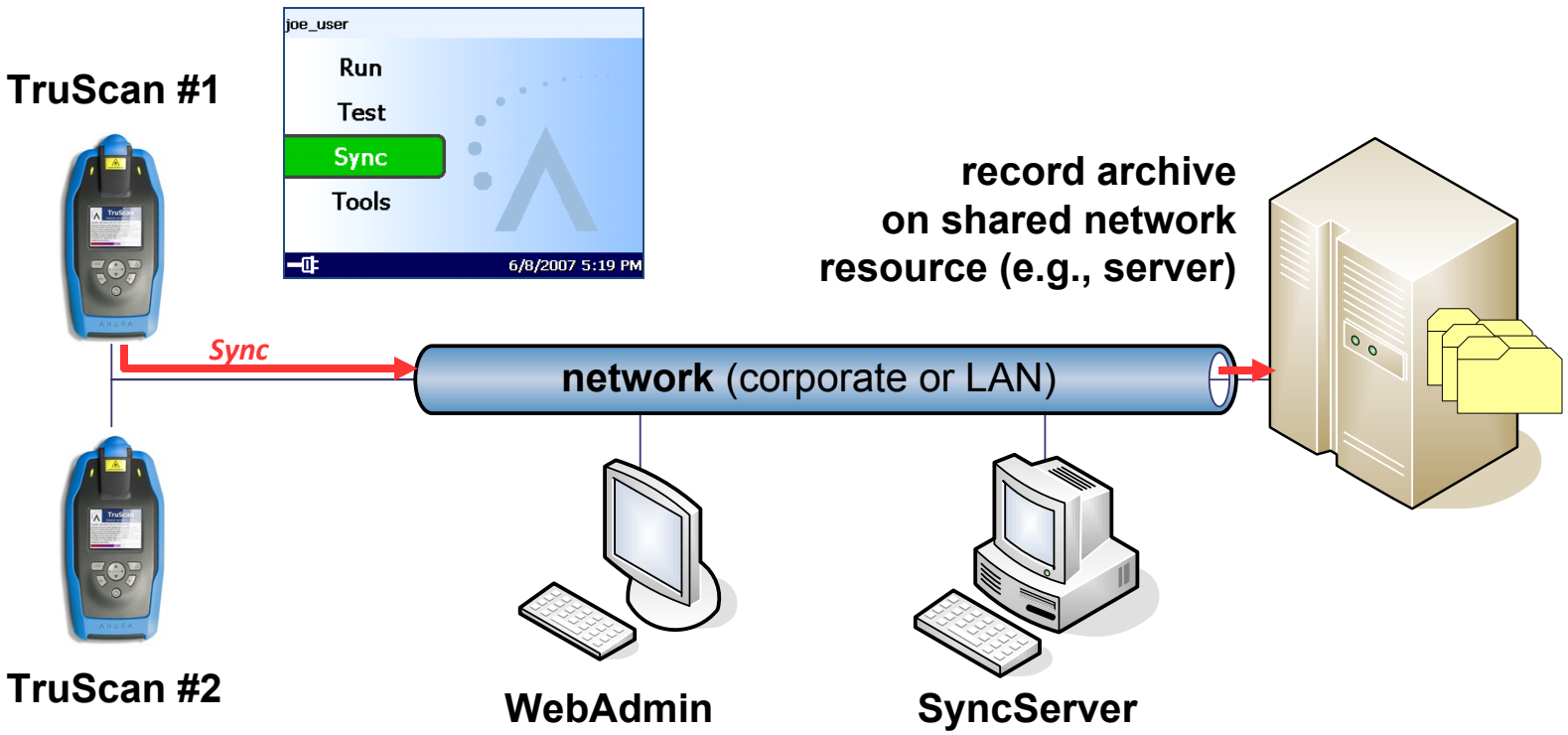
Fleet Management and Deployment of TruScan

- TruScan was designed to seamlessly manage inter-device method transfer, and fleet configuration

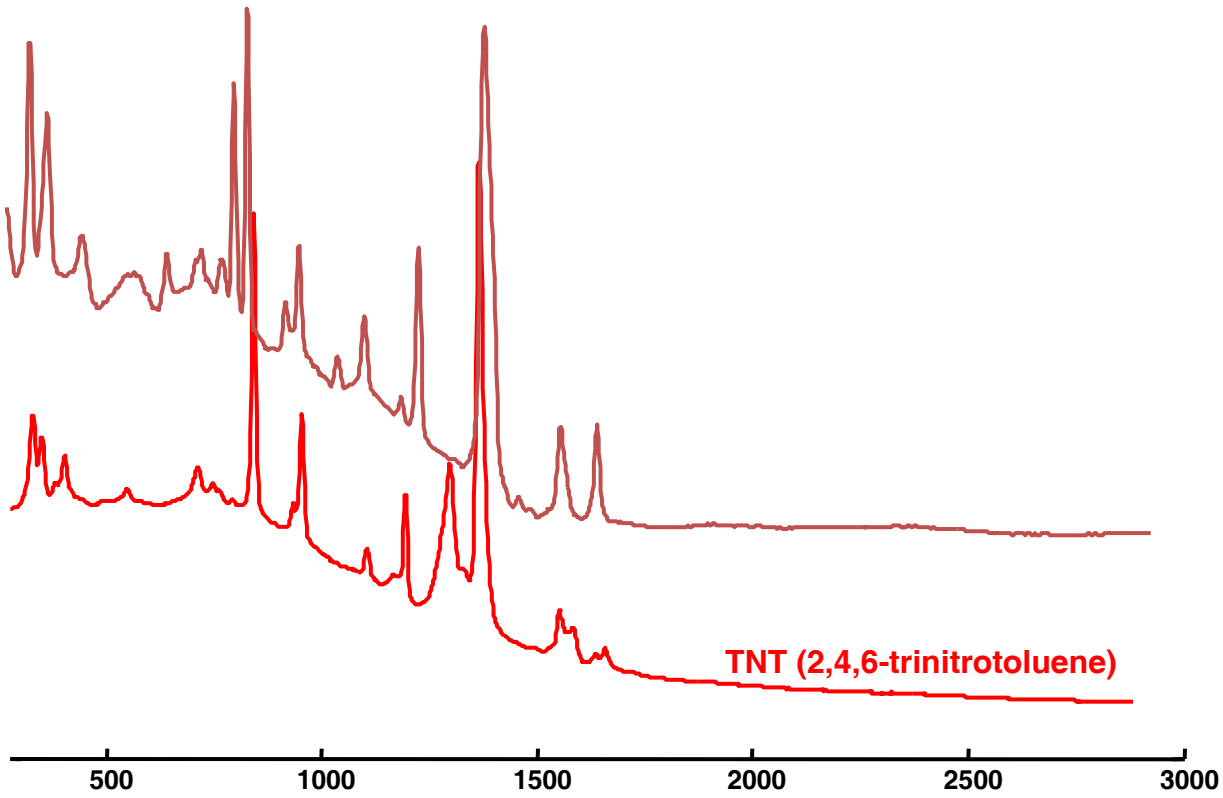
Inserting the CF-Ethernet Adapter

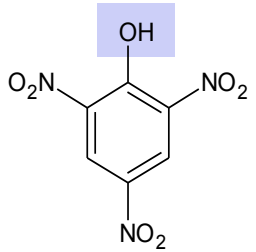


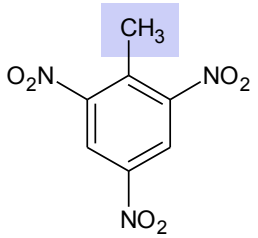
Data Archival



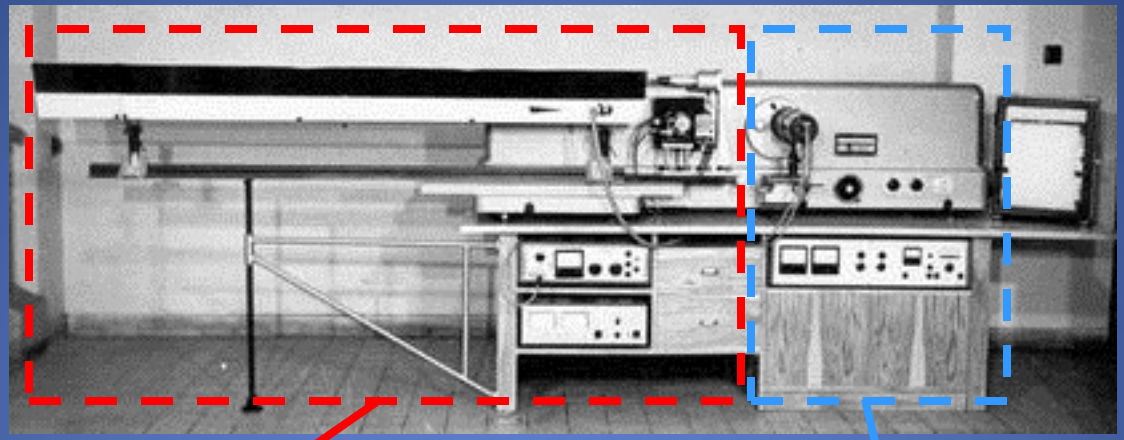
Functional Selectivity

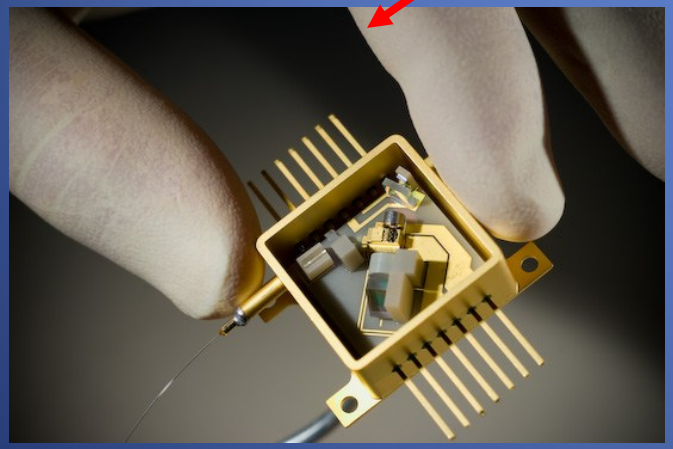


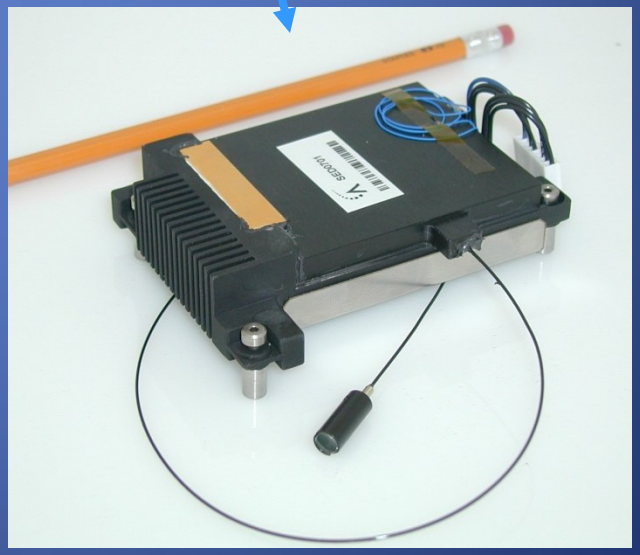




Progression of Raman Instruments



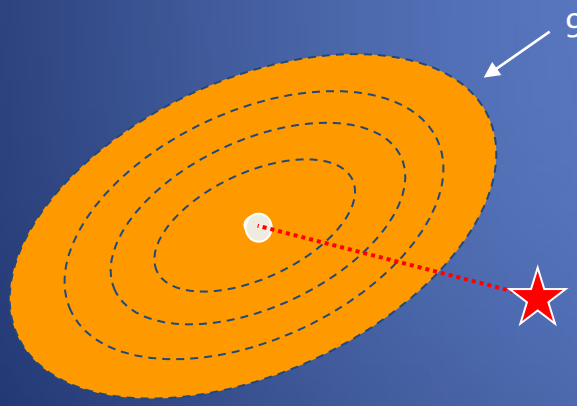




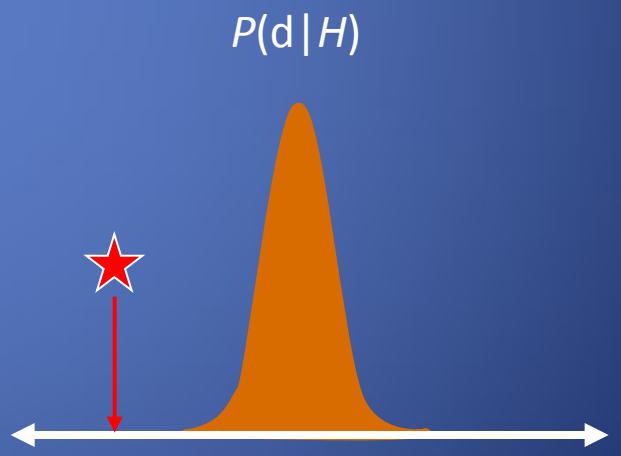
The Decision

Is the (multivariate) difference between the measured material and reference larger than we would expect given the uncertainty of the measurement? If YES, then FAIL.

On TruScan, this statistical decision is fully multivariate, using explicit knowledge of the uncertainty of the measurement at every channel on the detector, somewhat like a massively multivariate *t*-test.



multivariate illustration



univariate t-test

Robustnost systému

Jediný přístroj této kategorie, který splnil normu amády USA MIL-STD-810F.

Přístroj fungující v hloubce 1 m v nádrži

Přístroj fungující při teplotních cyklech

Vyhovuje normě MIL-STD-810F

TEST	Specifikace
Mechanický náraz	40g v 11ms, zub pily
Vibrace	1hr/axis, složené kruhové vibrace
Náraz při pádu	z výšky 122 m na překližkovou desku položenou na beton, 26 x
Vlhkost	5 x 48 hod, 60 °C a 95% relativní vlhkosti
Písek/prach/nečistota	foukání prachu
Teplotní šok	-30C až +60C,<1min
Nízká teplota (zapnuto)	-20C po dobu 1 dne
Vysoká teplota (zapn.)	+35C po dobu 3 dnů
Nízká teplota (skl.)	-30C po dobu 1 dne
Vysoká tepola (skl.)	+60C po dobu 7 dnů
Ponoření (zapnuto)	30 min. v hloubce > 1 metr