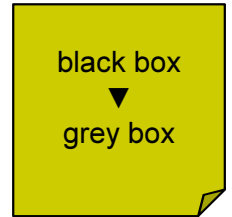


(Kép)pontról (kép)pontra: az infravörös képalkotás biomérnöki alkalmazásai



Gergely Szilveszter

Budapesti Műszaki és Gazdaságtudományi Egyetem

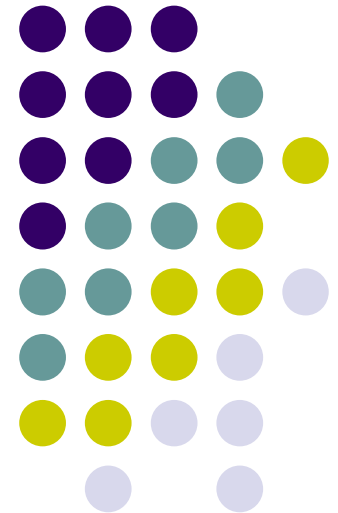
Alkalmazott Biotechnológia és Élelmiszertudományi Tanszék



M Ű E G Y E T E M 1 7 8 2

PerkinElmer Szeminárium

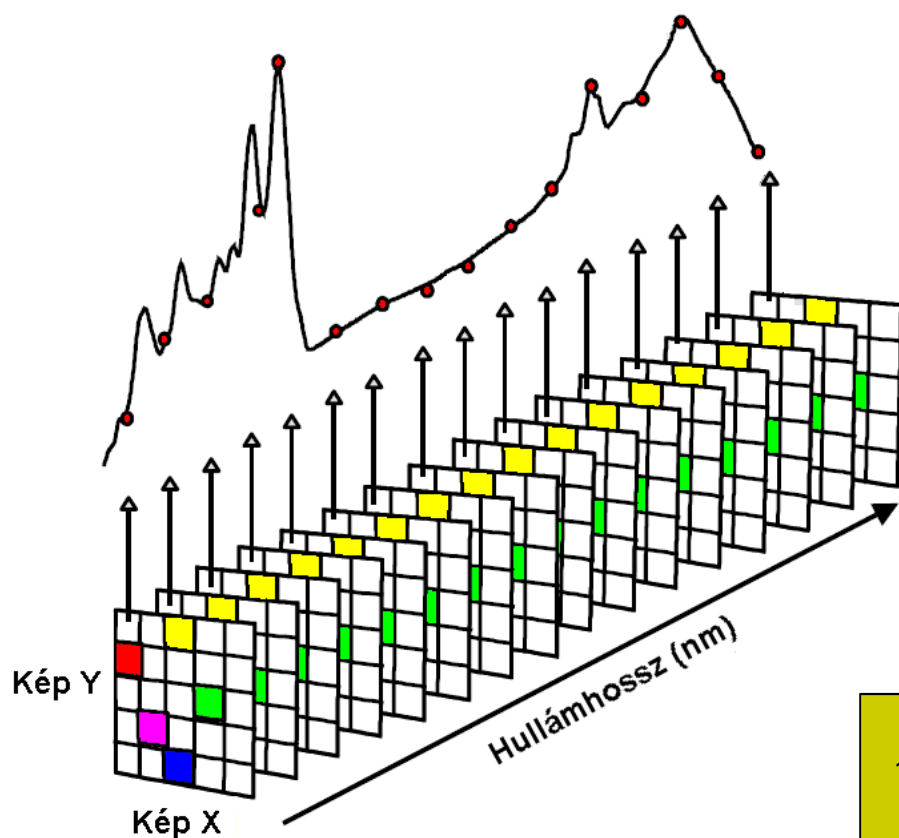
2015. október 20.





Spektrális hiperkocka vs. tudomány

- Spektrális hiperkocka



De mi volt
1994 előtt?

Gyilok.

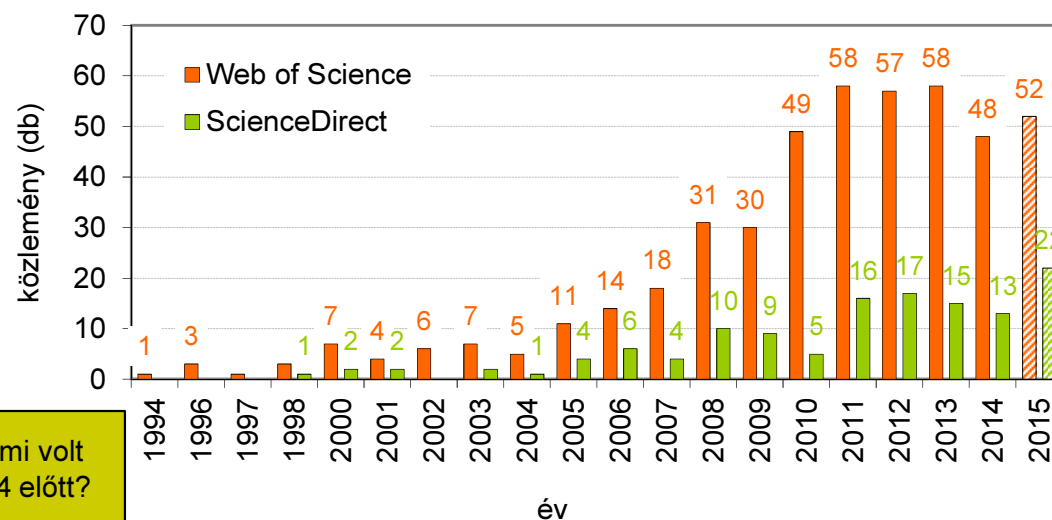
- „near-infrared AND imaging
AND pharma” kulcsszóra
történő keresés

- Web of Science

- ScienceDirect



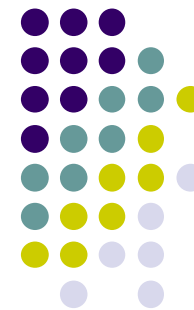
- Progresszív fejlődés



Lewis E.N., Carroll J.E., Clarke F.:

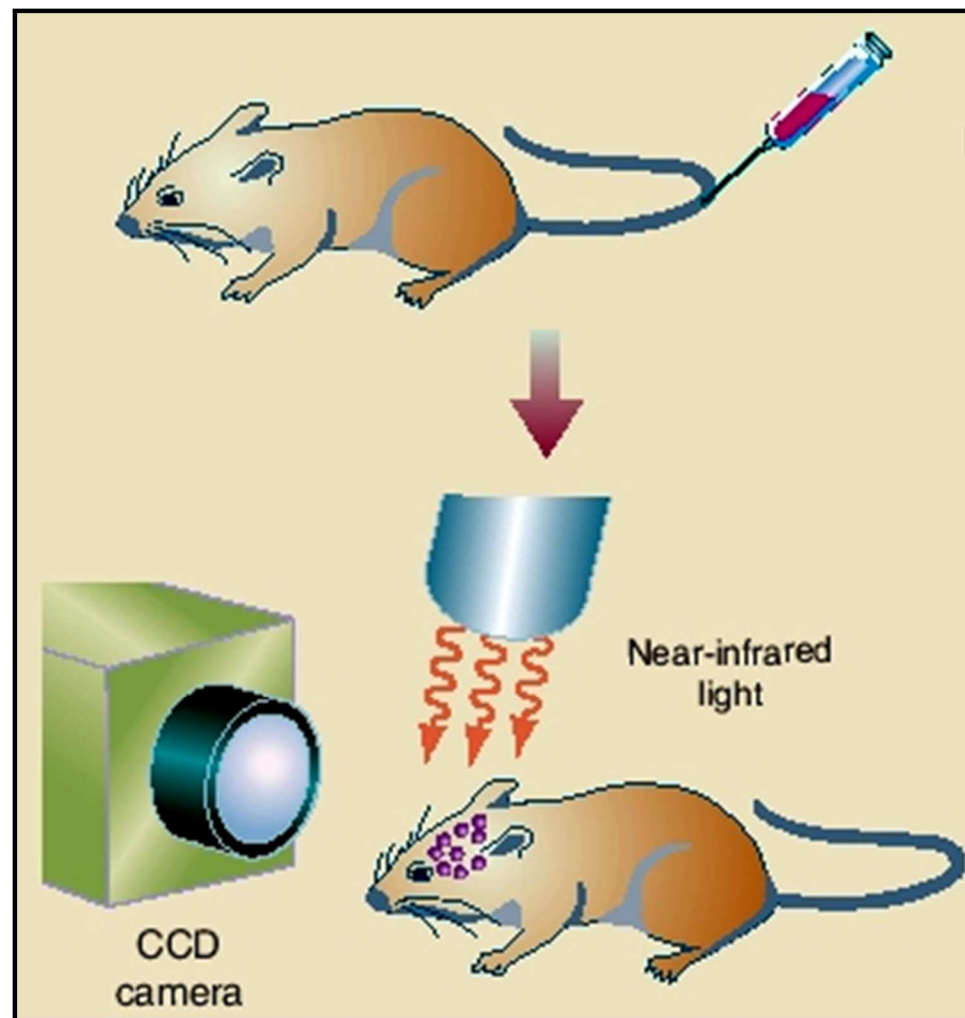
A near infrared view of pharmaceutical formulation analysis.

NIR news 12(3), 16–18 (2001)



Egér a Marsról – az elv...

Itt már aktív:
IR besu-
gárzás, DE
roncsolás-
mentes



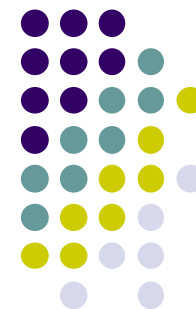
Sokszor csak
a bonc-
asztalon látjuk
az Alzheimer-
kór állapotát.

Korai észlelés
és terápia:
jobb esélyek.
NIR kamera
segít.

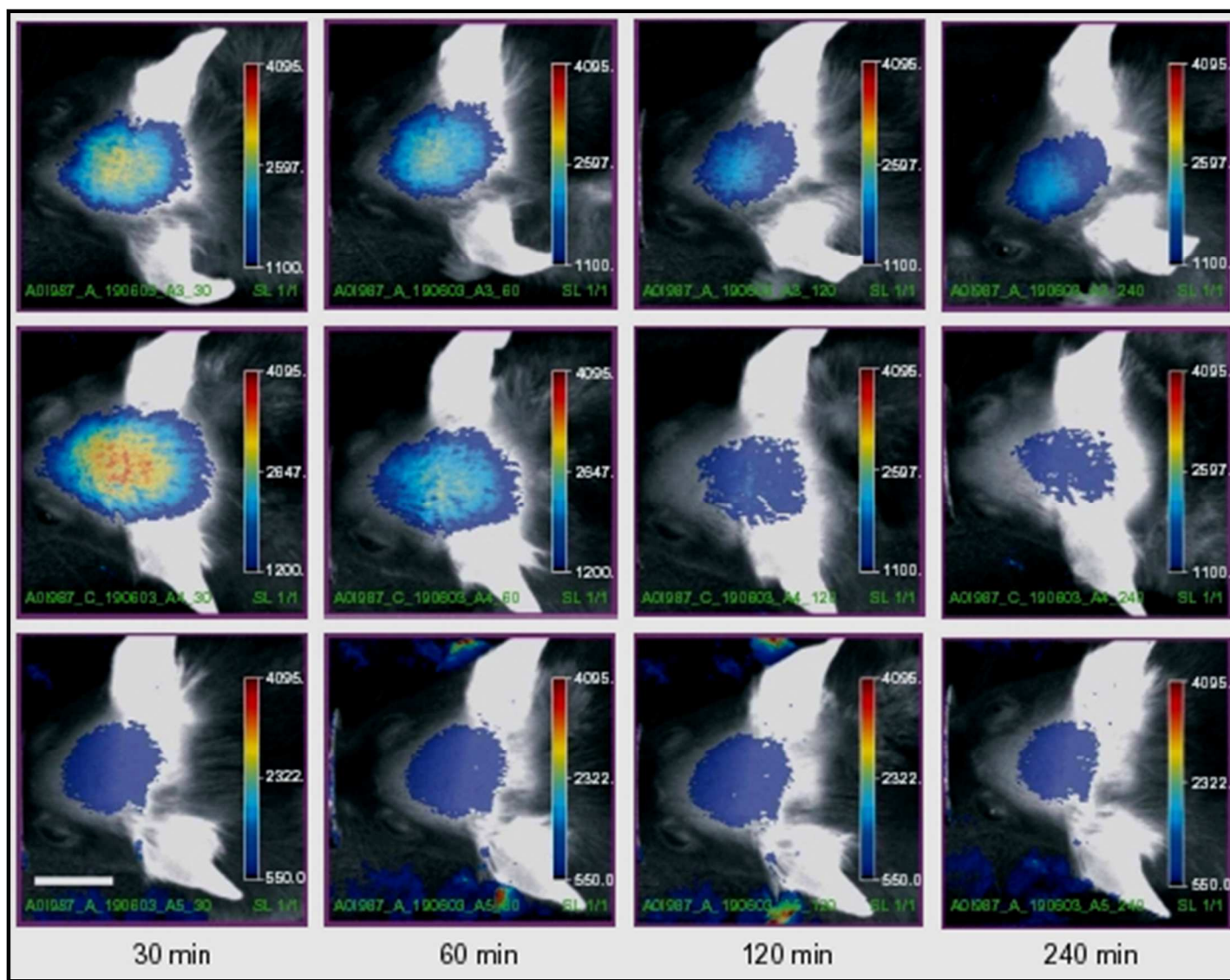
Golde T.E., Bacskai B.J.:

Amyloid deposits can be rapidly detected in the brains of living mice using a novel ligand and near-infrared fluorescence imaging.

Nat. Biotechnol. 23(5), 552–554 (2005)



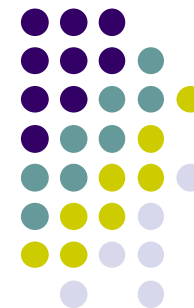
Egér a Marsról – ... és a gyakorlat



Hintersteiner M. *et al.*:

In vivo detection of amyloid- β deposits by near-infrared imaging using an oxazine-derivative probe.

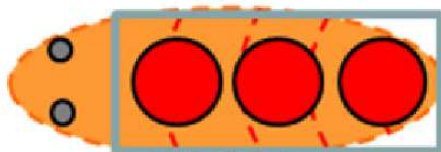
Nat. Biotechnol. 23(5), 577–583 (2005)



Selyemhernyó szexálása – az elv, ...

TOP VIEW

Silkworm Pupa



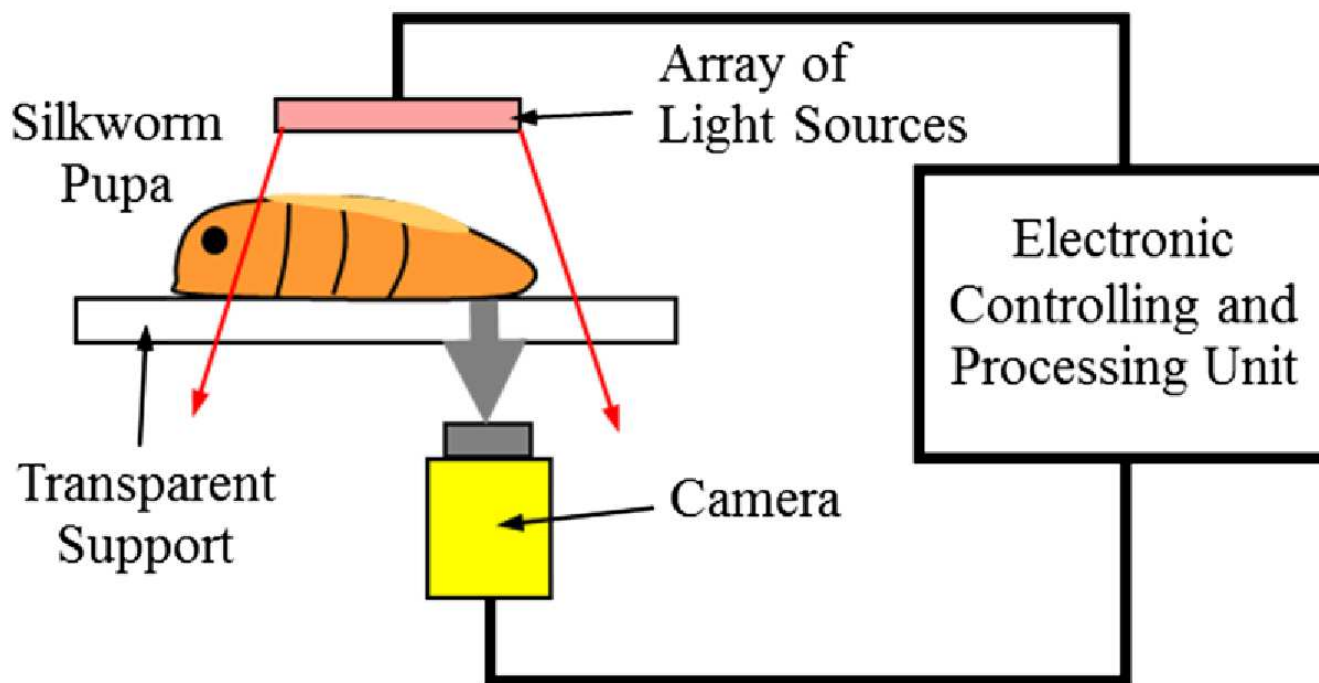
Array of Light Sources

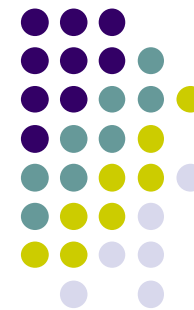
Tenyésztés szempontjából fontos, hogy mi van a bábban.

Bábot megbontani nem lehet, mert akkor elpusztul...

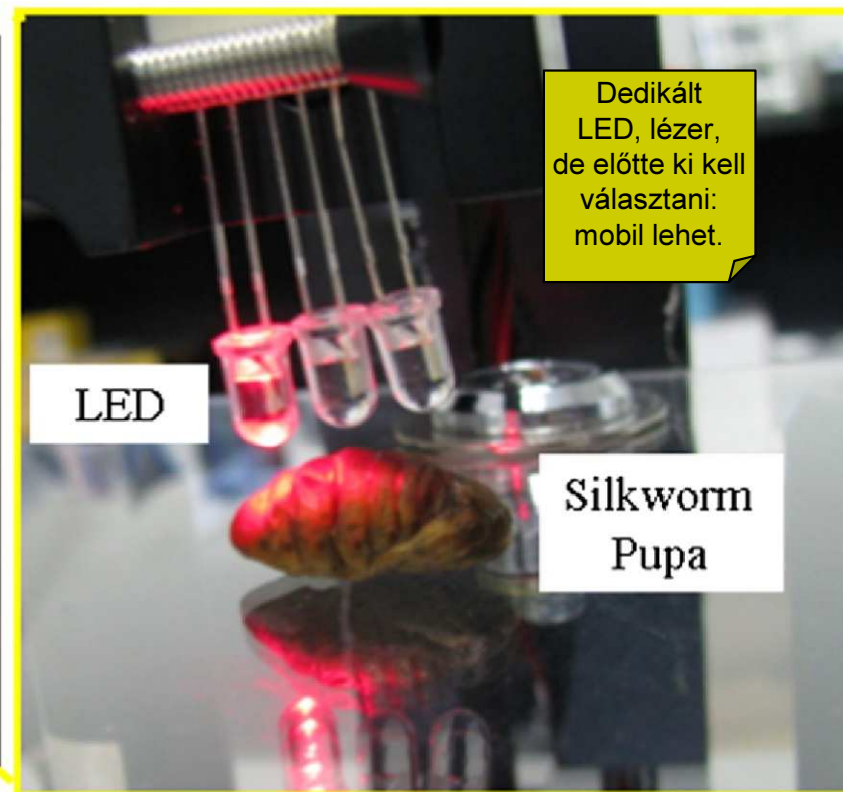
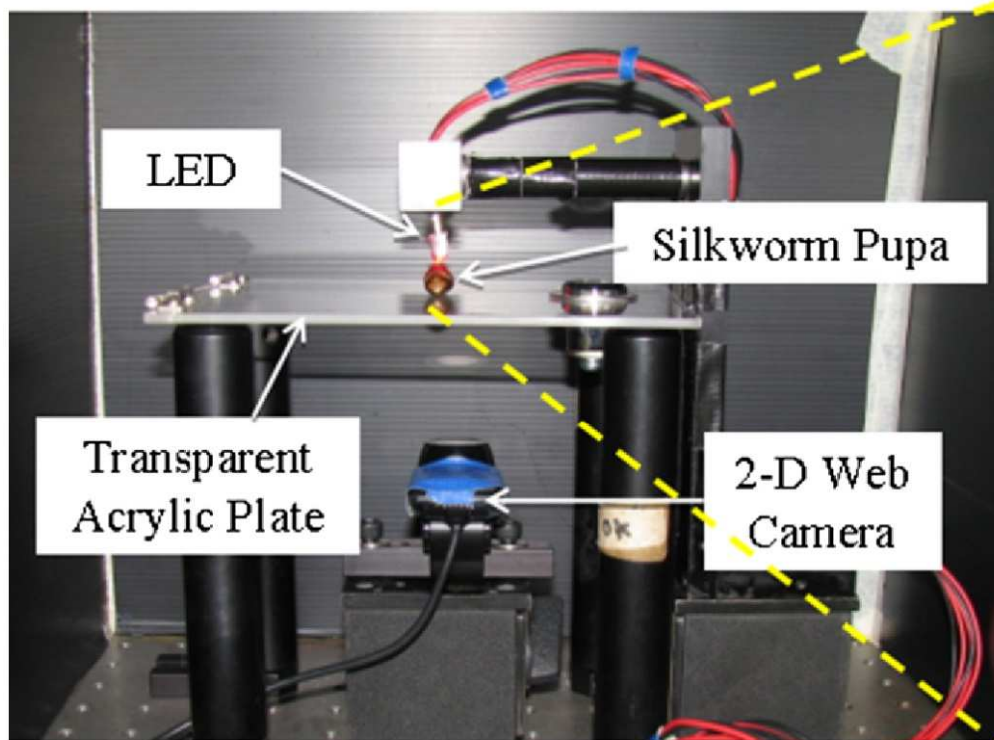
Nézzünk bele a NIR segítségével a selyemhernyó bájába!

SIDE VIEW

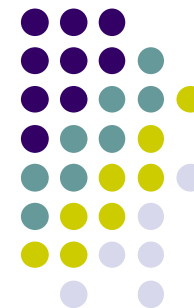




Selyemhernyó szexálás – ... a „vas” ...



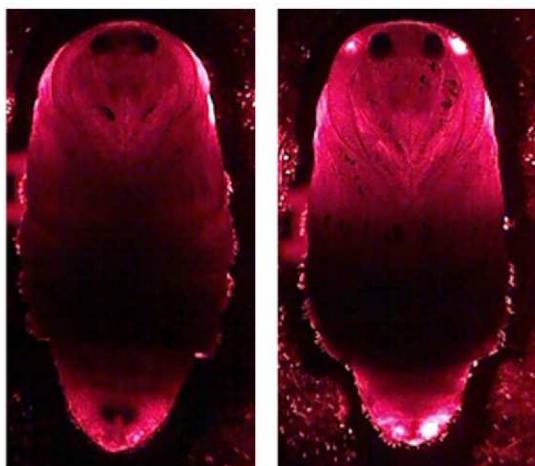
Close Up View



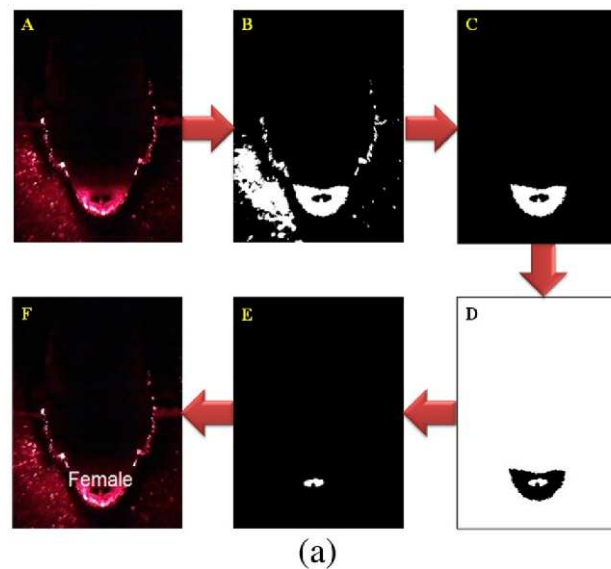
Selyemhernyó szexálás – ... és a kimenet



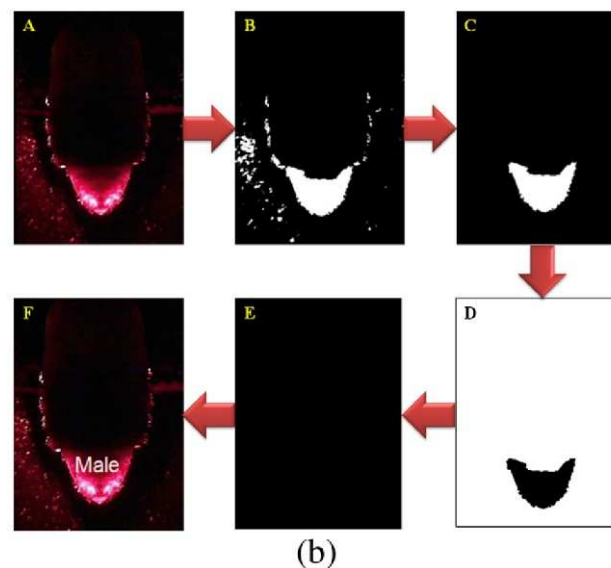
(a)



(b)

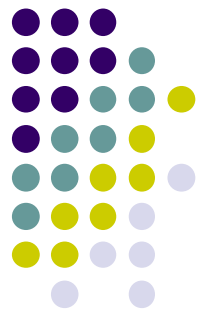


Lányoknál
semmi
extra ...



... de a fiúknak
világít
a „farkuk”.

Vénaszkenner



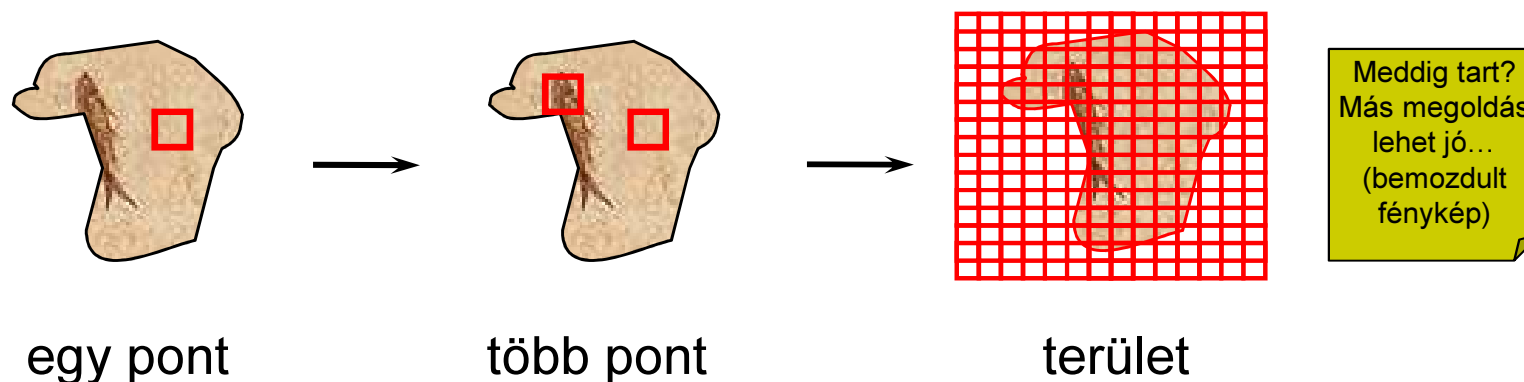
NIR 2007
Umeå (S):
papírra
„H₂O”

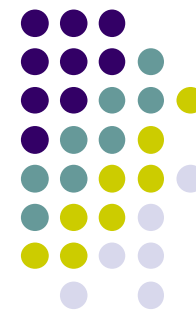
Mozizzunk?

Mikro/makroszkópos képalkotás – fő pontokban

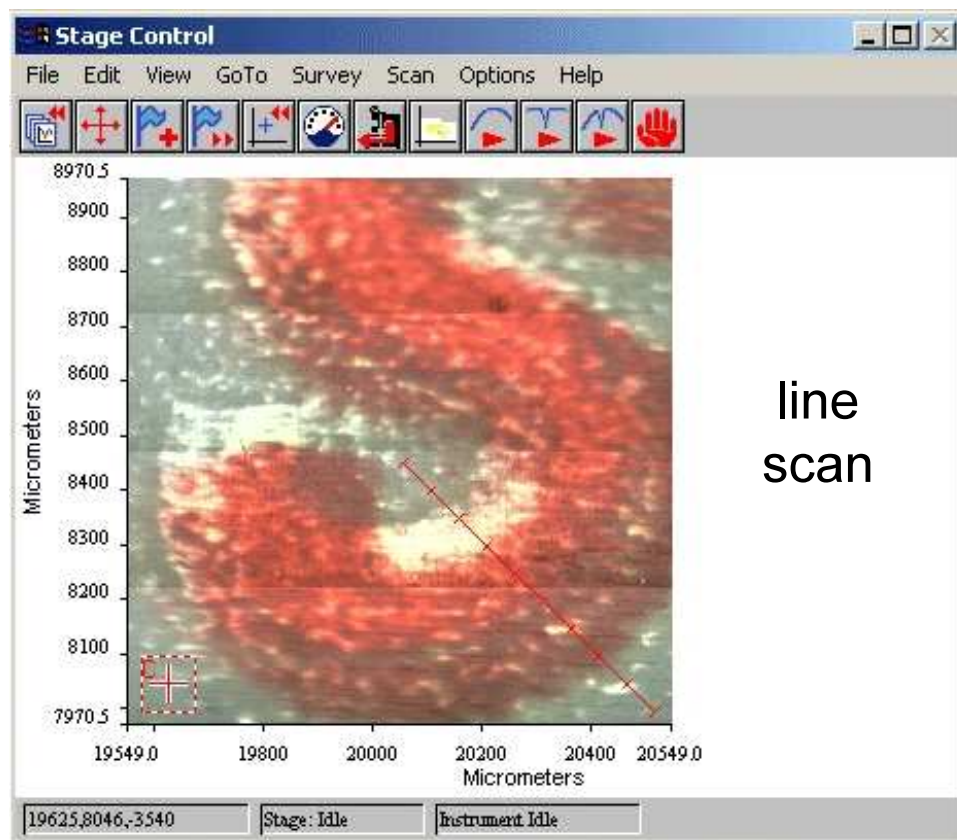


- Pár/tíz mikrométeres nagyságrendek(től) pár centiméterig
- Látható kép: vizsgálandó terület megkeresése, majd (N)IR mérés
- Nem, vagy csak „kis” mintaelőkészítést igényel: rutin...
- Különböző mérési módok: heterogén minták könnyebb vizsgálata

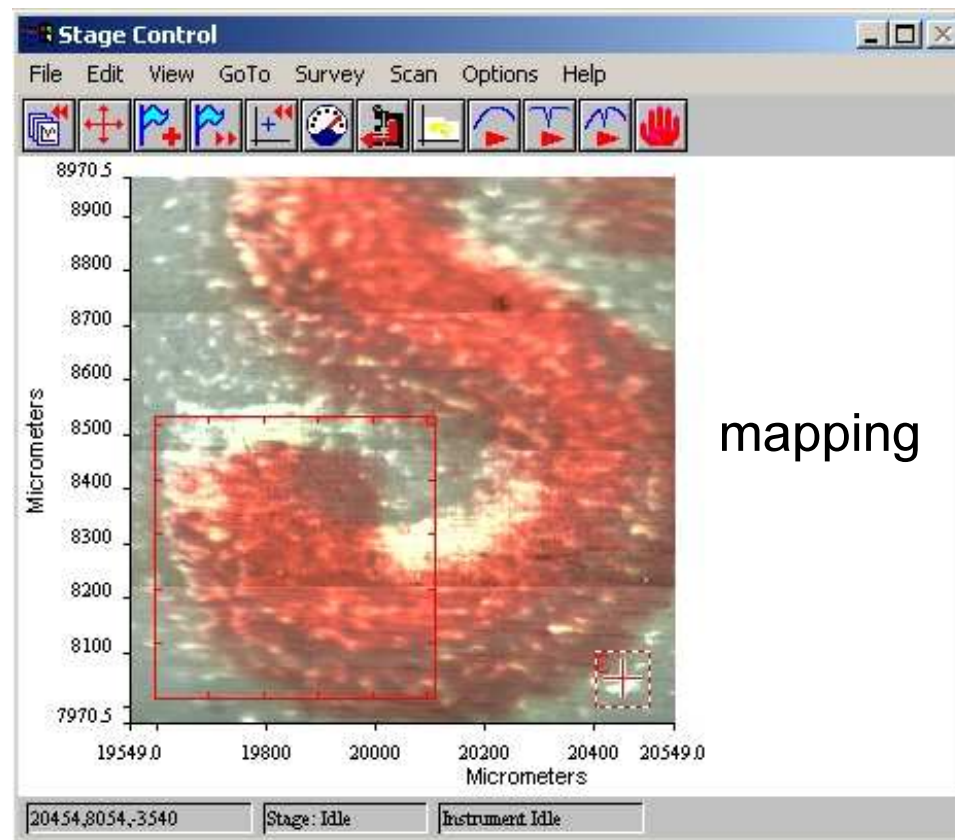




Mikro/makroszkópos képalkotás – sok pont = vonal (1D), térkép (2D)



line
scan

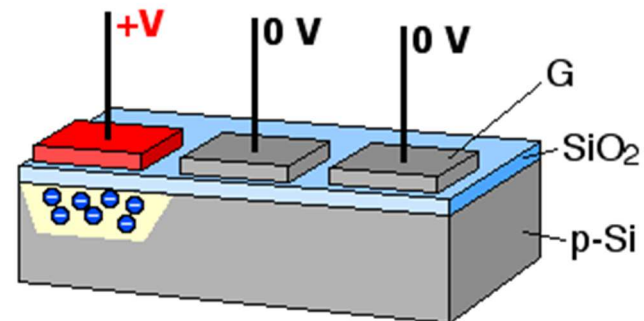


mapping

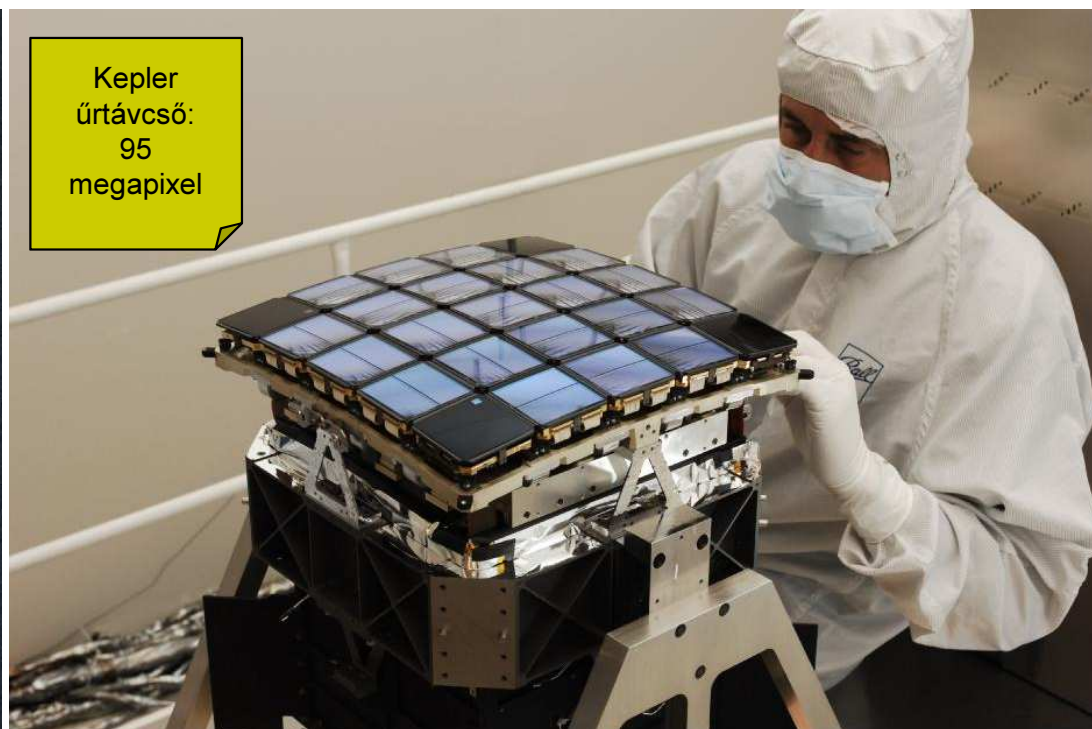
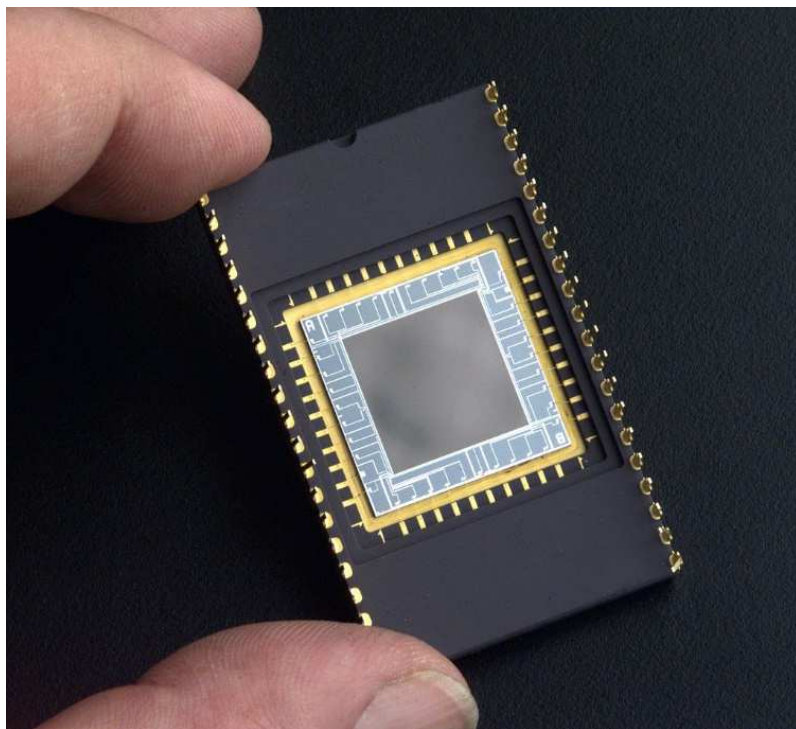


Mikro/makroszkópos képalkotás – FPA

- FPA (Focal Plane Array)



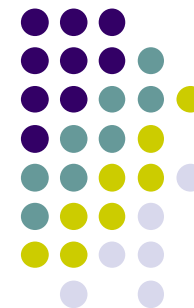
videotelefon
fejlesztése
(AT&T): 1969!



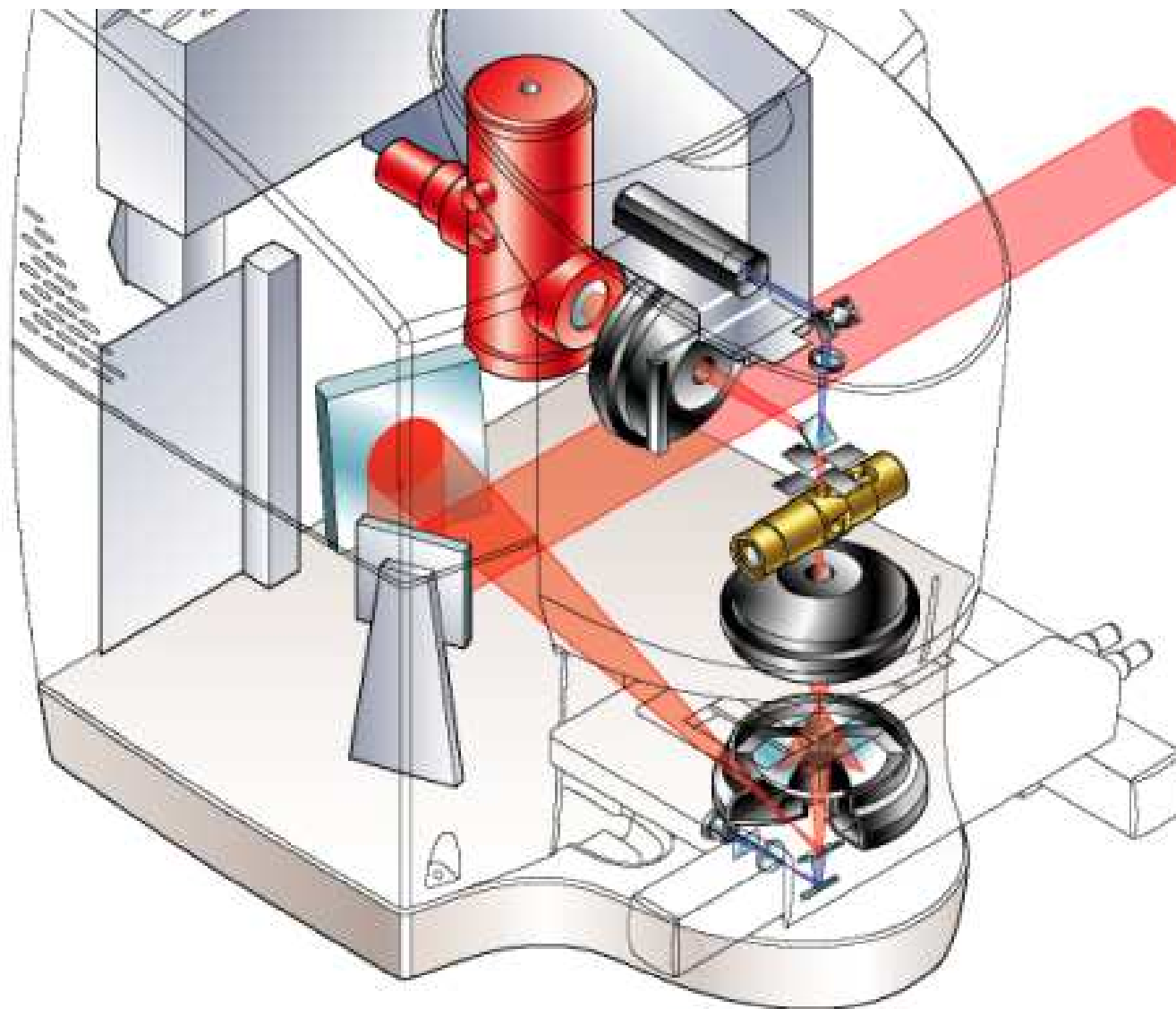
Kepler
űrtávcső:
95
megapixel

https://en.wikipedia.org/wiki/Charge-coupled_device

http://www.nasa.gov/mission_pages/kepler/multimedia/images/kepler-focal-plane-assembly.html



Mikro/makroszkópos képalkotás – T

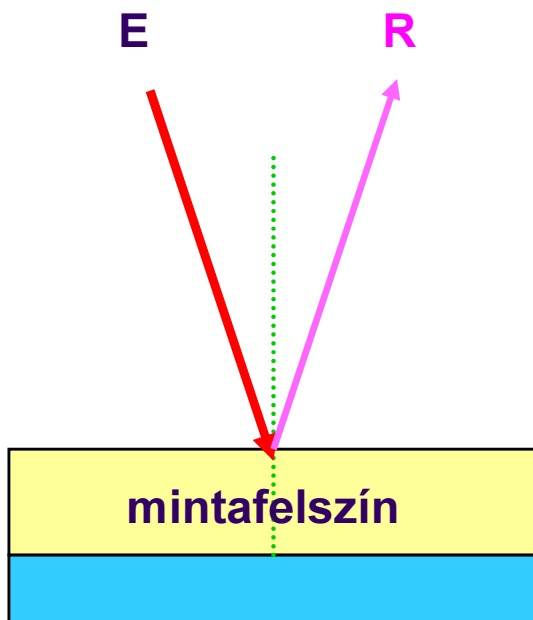


T
(transz-
misszió)

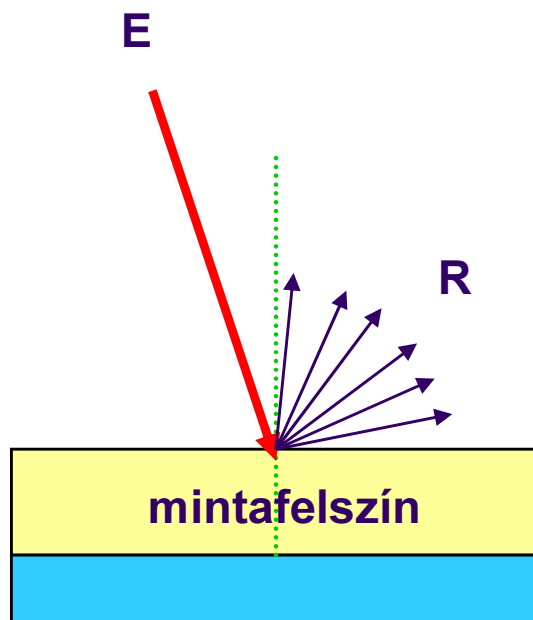


Mikro/makroszkópos képalkotás – R-ek

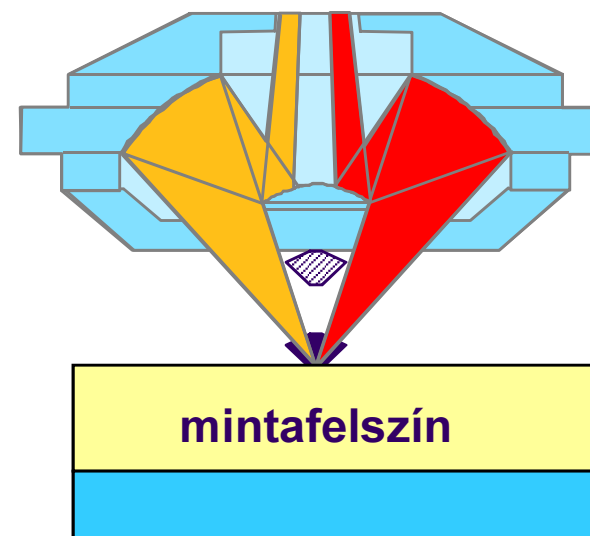
spekuláris
reflexió



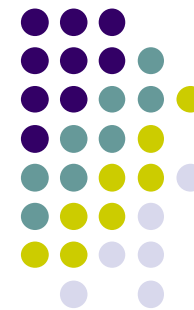
diffúz
reflexió



gyengített
teljes reflexió
(ATR)



R
(reflexió)

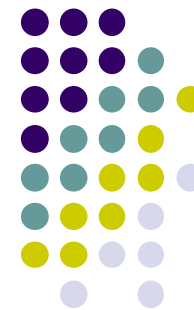


Mikro/makroszkópos képalkotás – Ge

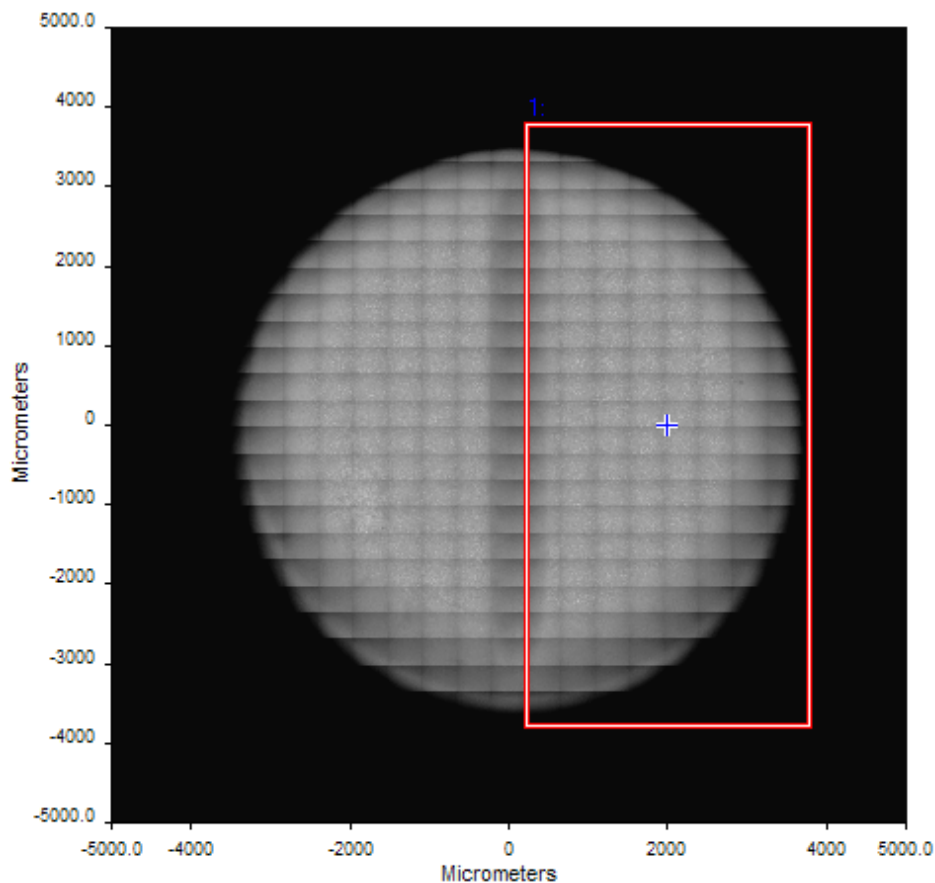


Eredetiségvizsgálat vs. imaging: makro (VIS)

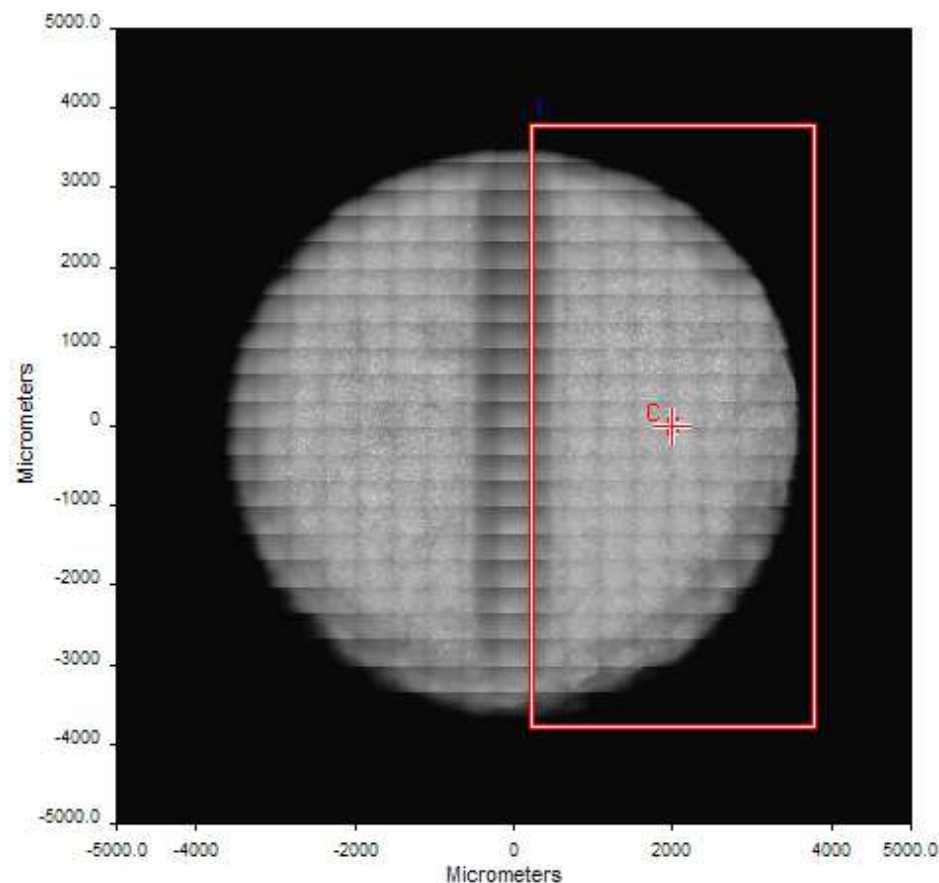
Edzőteremben
izomnövelő
helyett
„barkács
cucc”.



- Eredeti

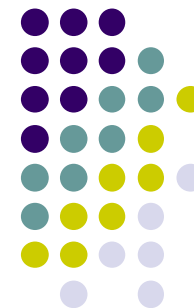


- Hamis



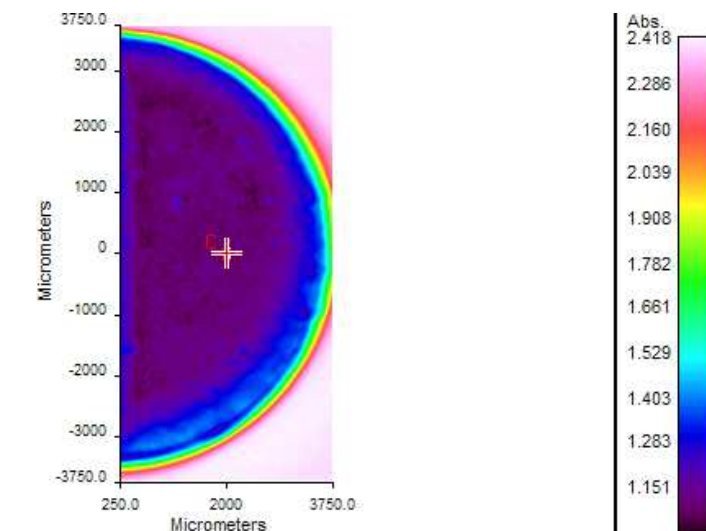
Eredetiségvizsgálat vs. imaging: makro (NIR)

mátrixhatás

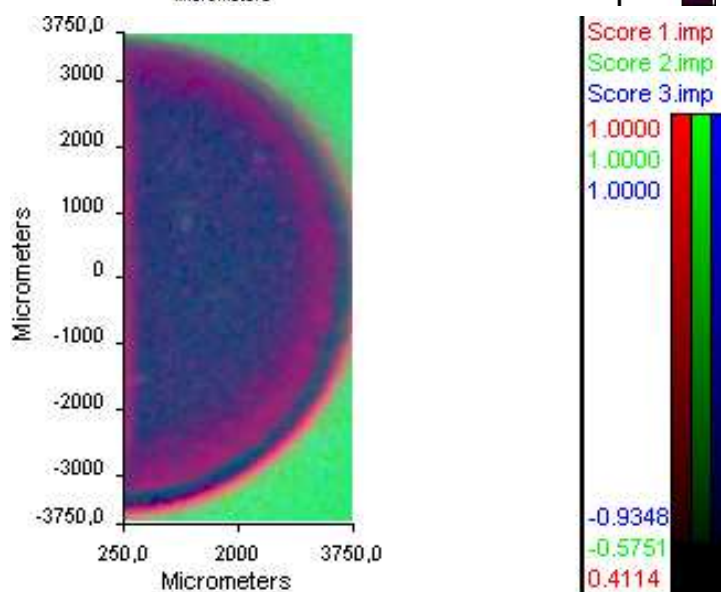


- Eredeti

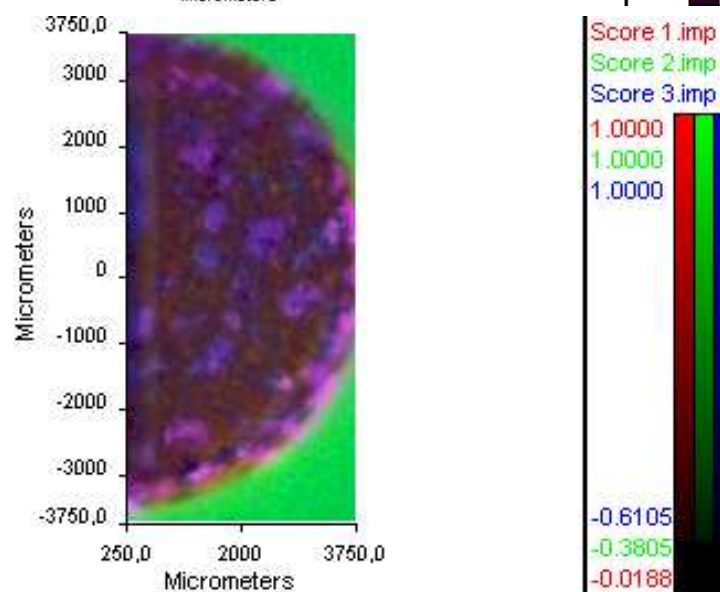
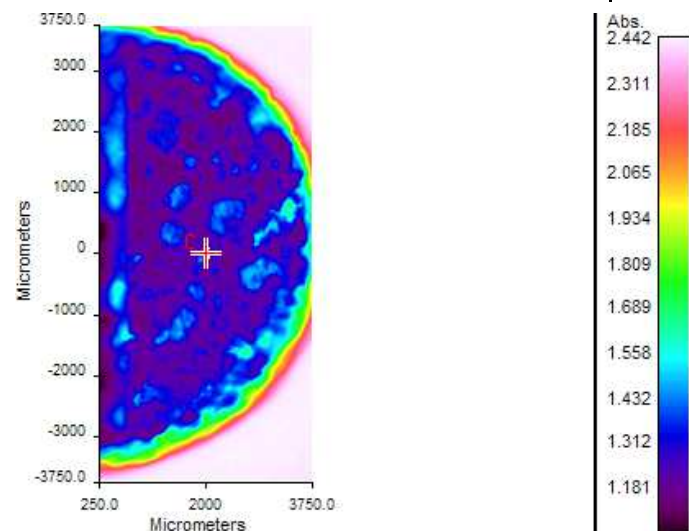
Abs.

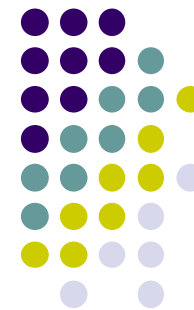


PCA

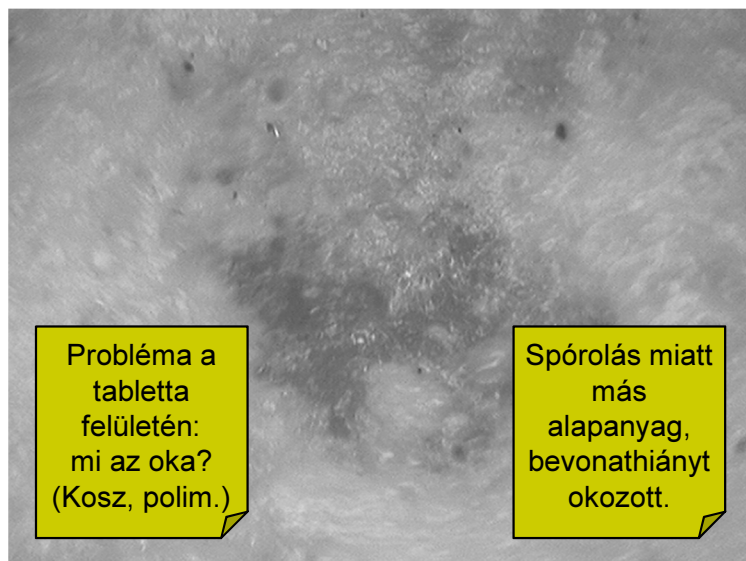


- Hamis

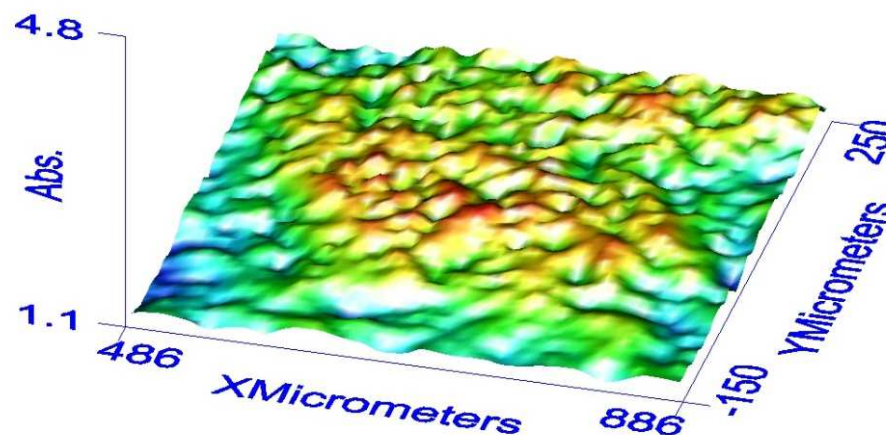
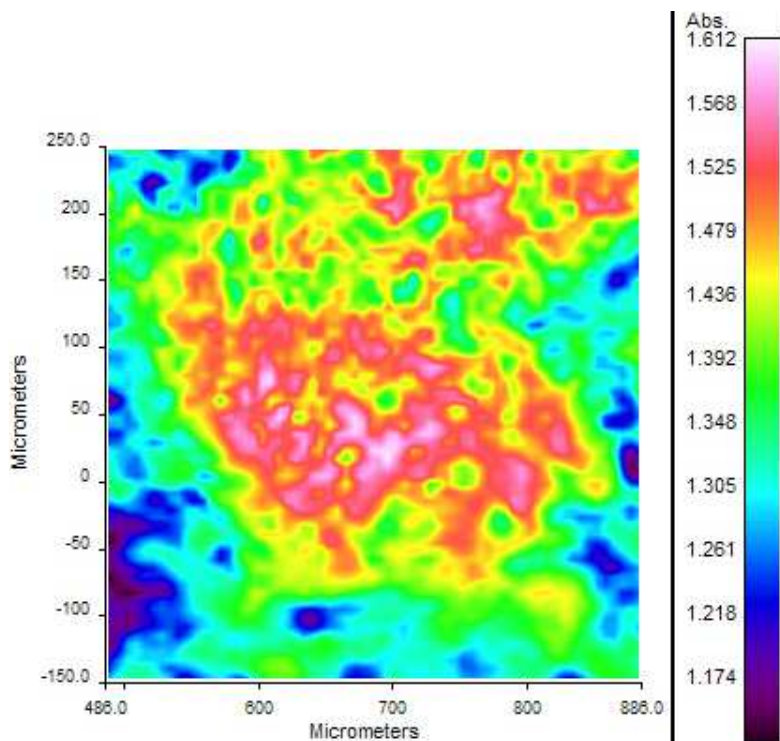




Technológiai probléma vs. imaging I.



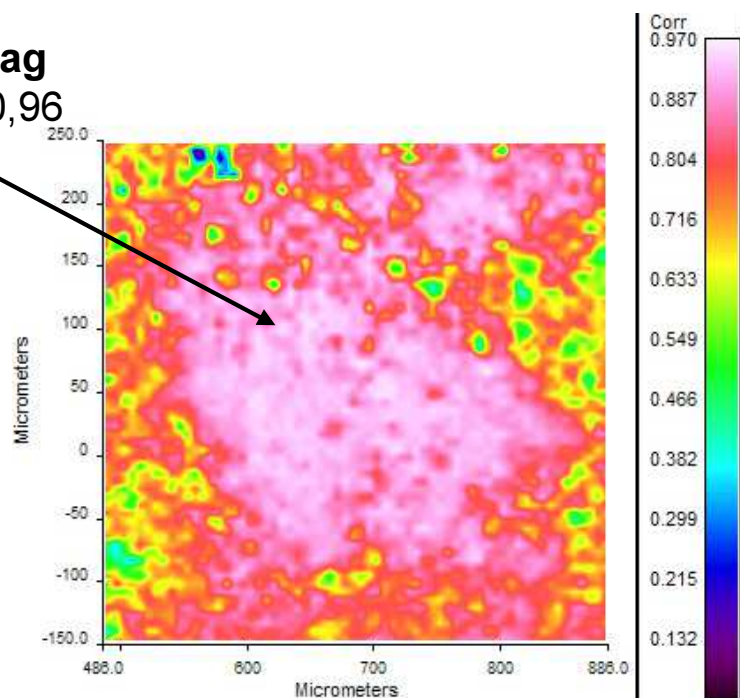
- Képkalkotás NIR tartományban: nincs roncsolás
- $400 \times 400 \mu\text{m}$ méretű mérési tartomány, $6,25 \times 6,25 \mu\text{m}$ méretű képpontokkal
- 64×64 pont, azaz összesen 4096 pont, pontonként 128 spektrum átlagolásával
- Átlagos abszorbanca értékek alapján színezett ábrák



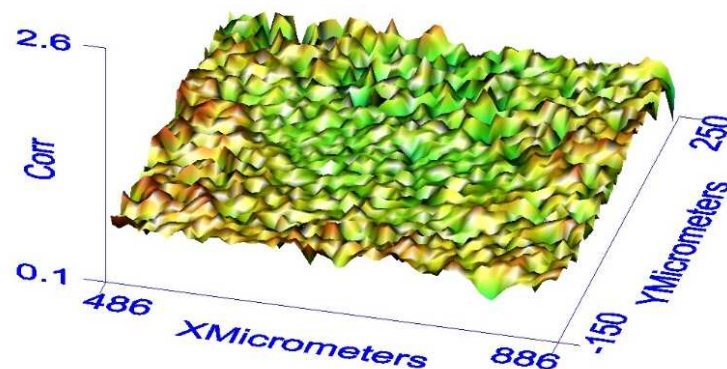
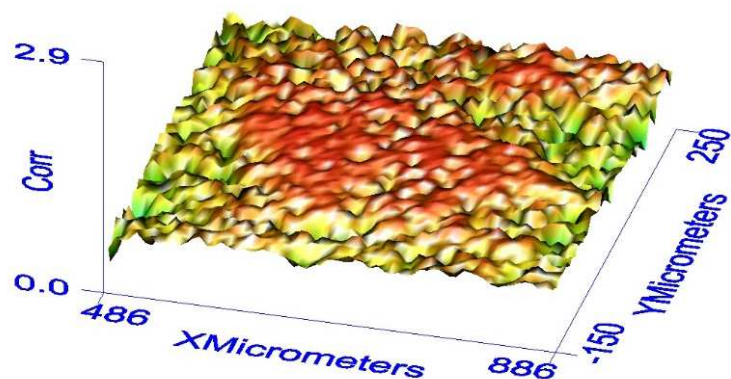
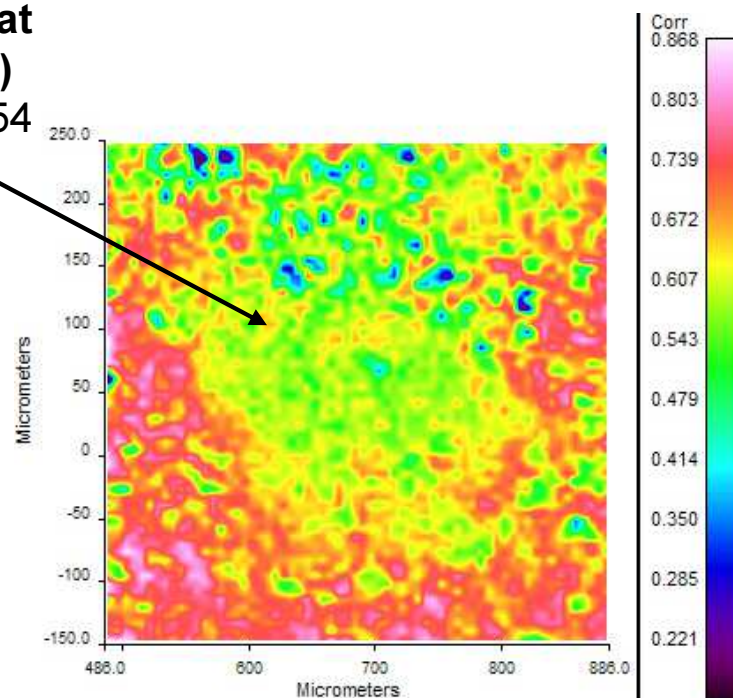


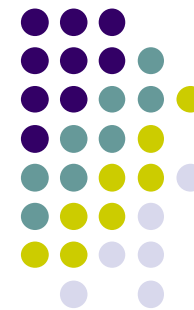
Technológiai probléma vs. imaging I.: korrelációs térképek (2D és 3D)

Hatóanyag
 $R^2=0,88-0,96$



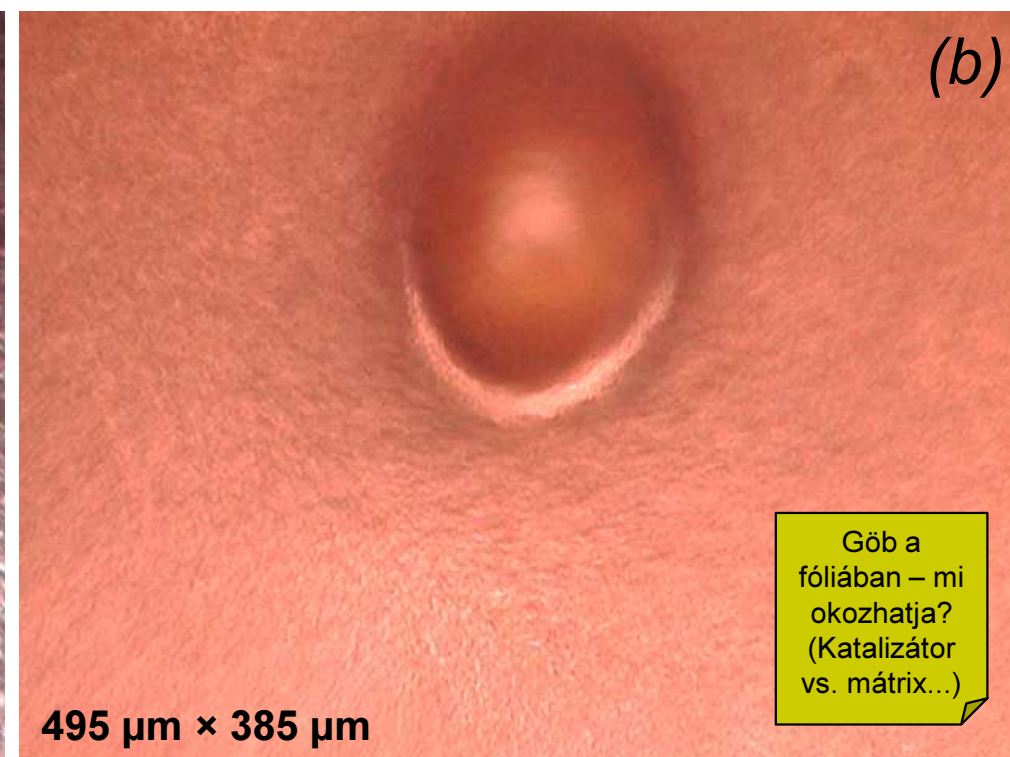
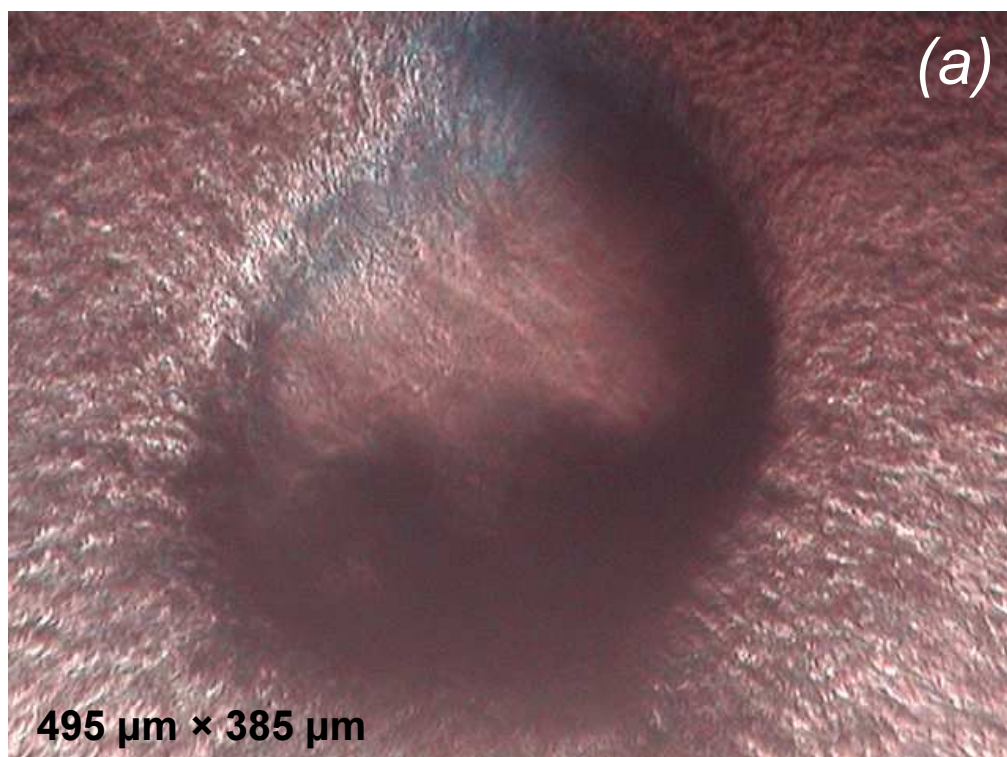
**Filmbevonat
(Opadry®)**
 $R^2=0,41-0,54$

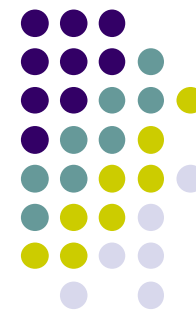




Technológiai probléma vs. imaging II.

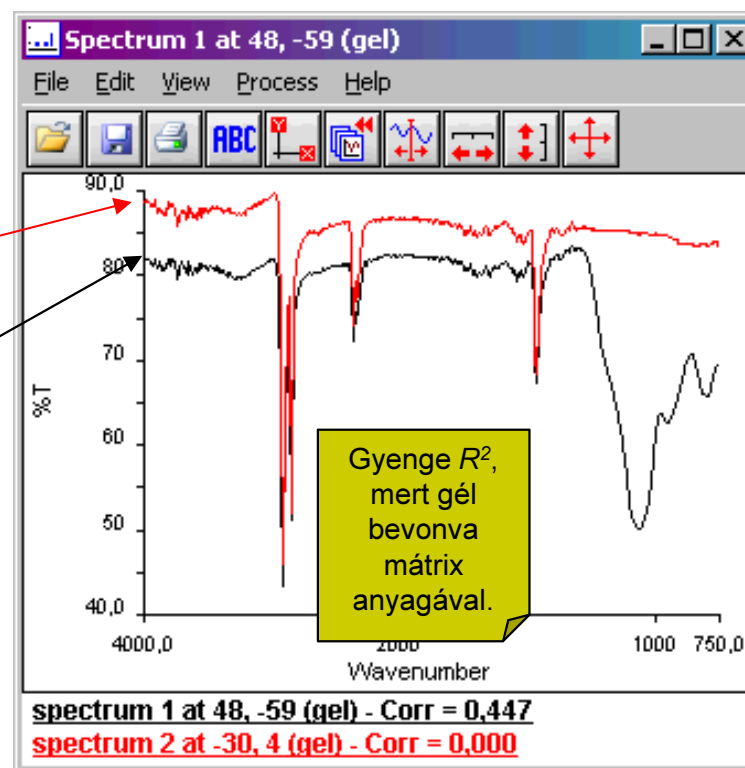
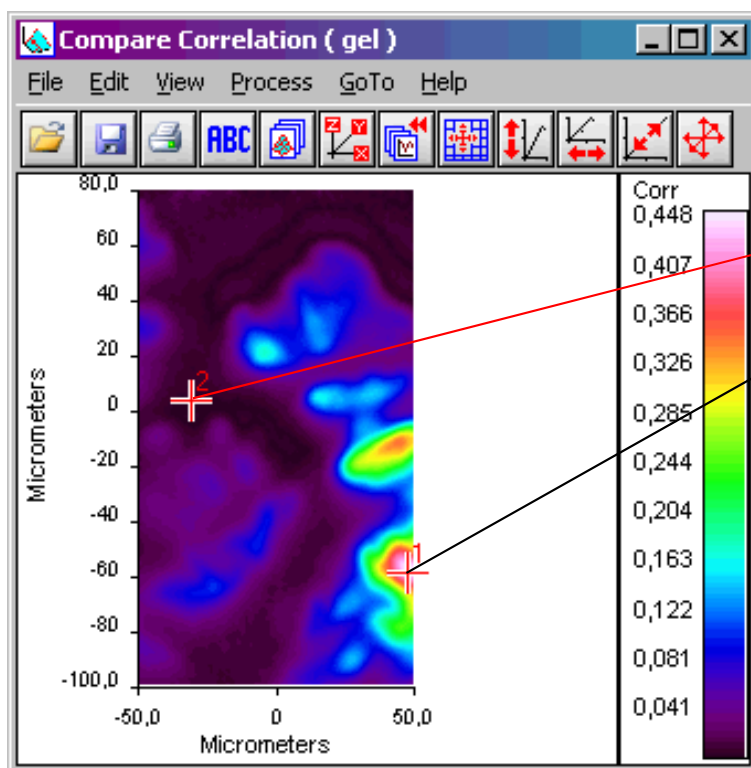
- transzmissziós kép (a) a transzmissziós, ill. reflexiós kép (b) a mikro ATR felvétel előtt



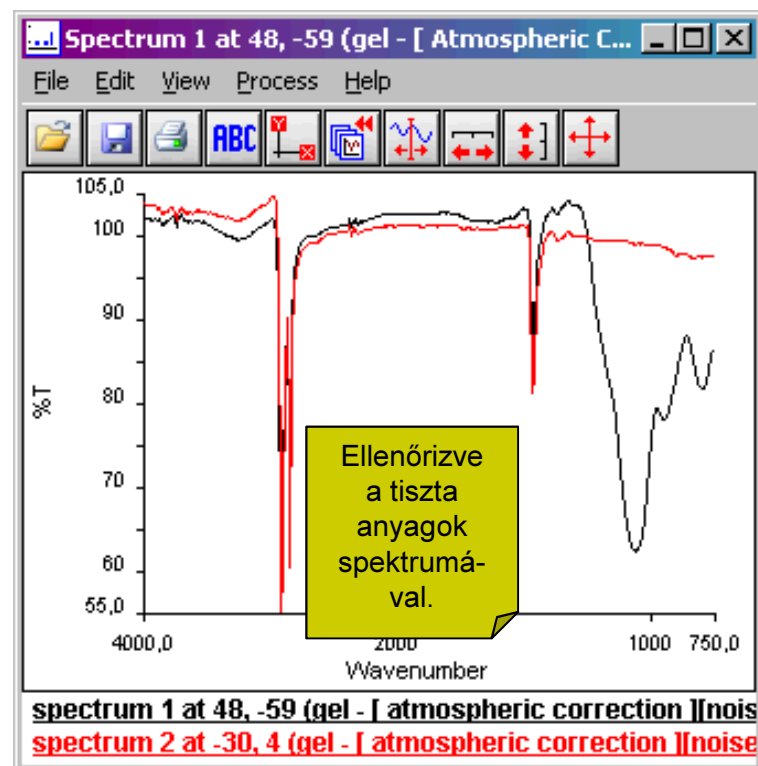
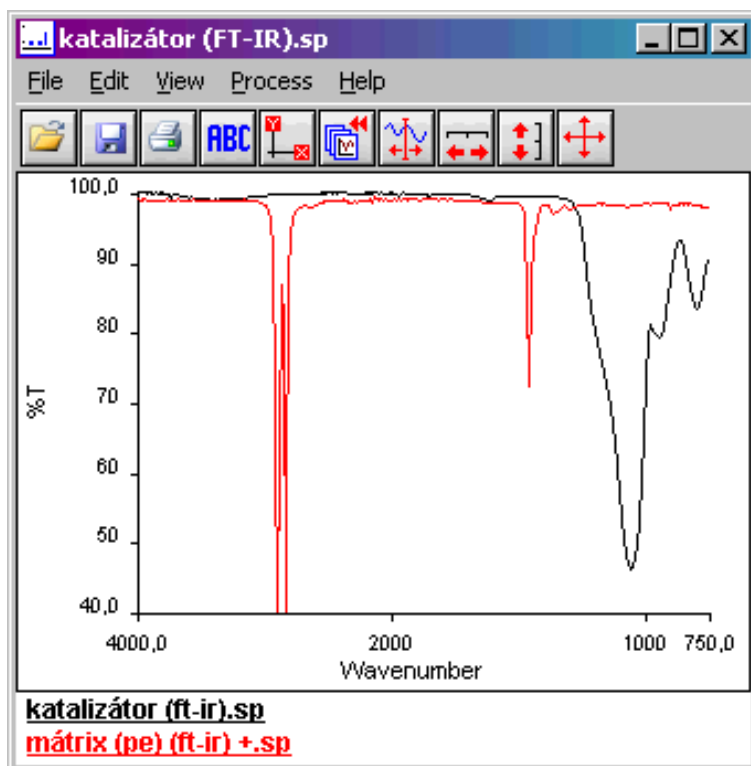
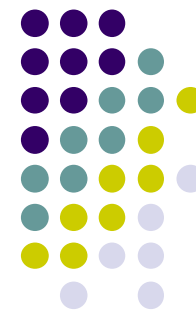


Technológiai probléma vs. imaging II.: korrelációs térkép: min. vs. max. R^2

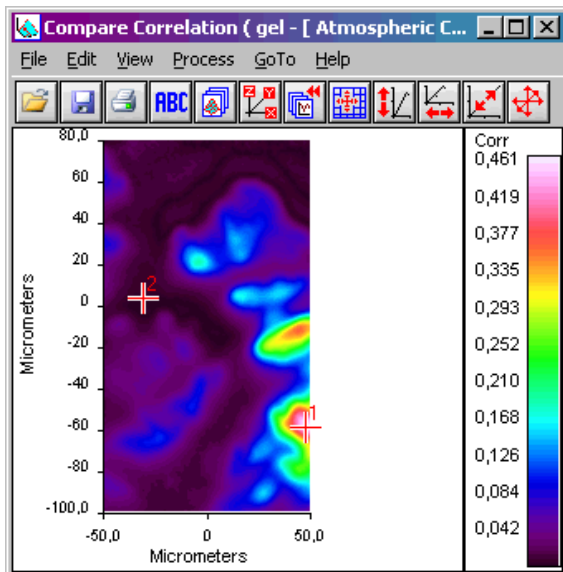
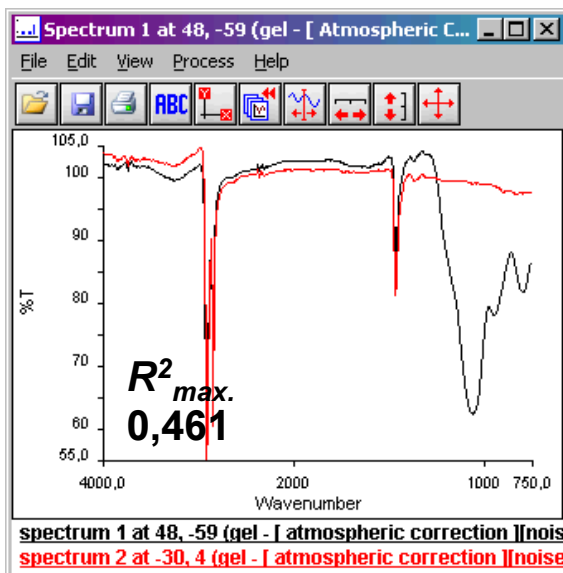
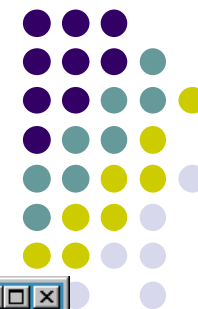
- R^2 (min.) = 0,000 : képen **2** (-30;4) ► **spektrum 2** —
 R^2 (max.) = 0,447 : képen **1** (48;-59) ► **spektrum 1** —



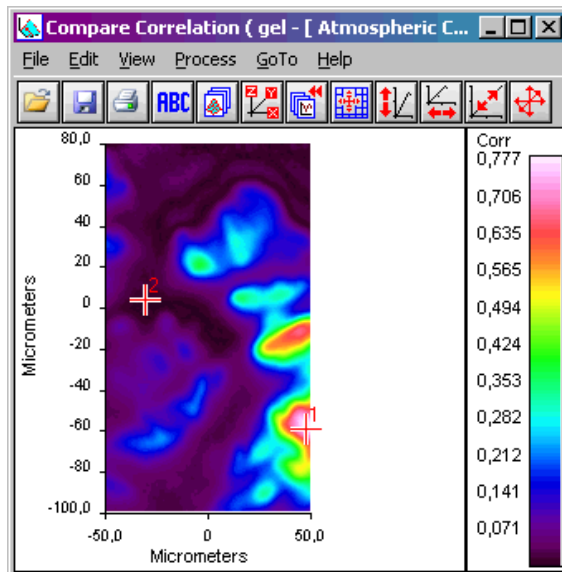
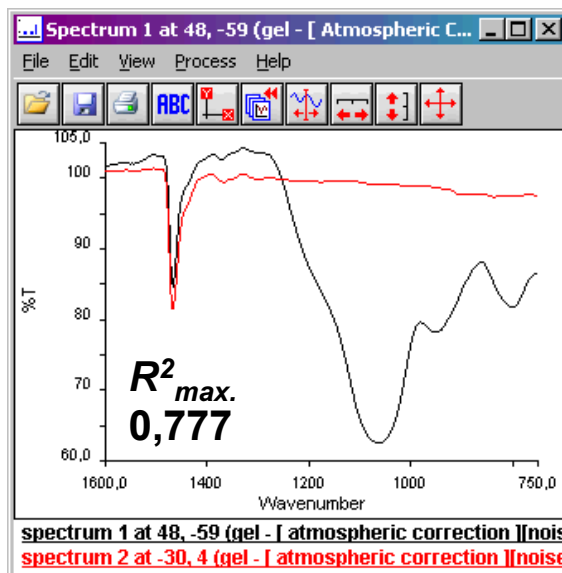
Technológiai probléma vs. imaging II.: Makro ATR (UATR) vs. mikro ATR (im.)



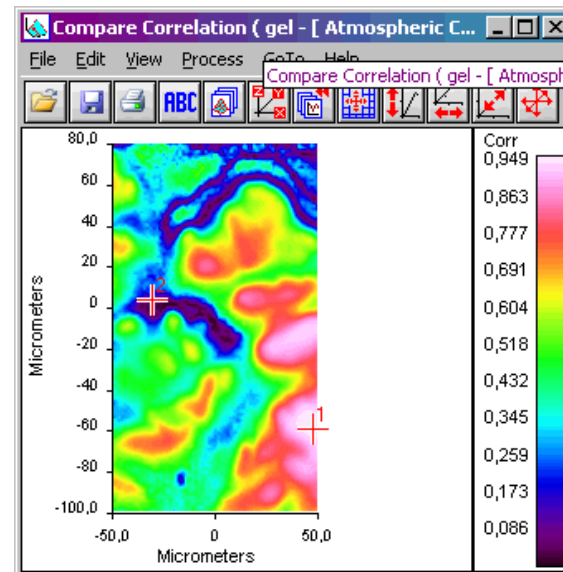
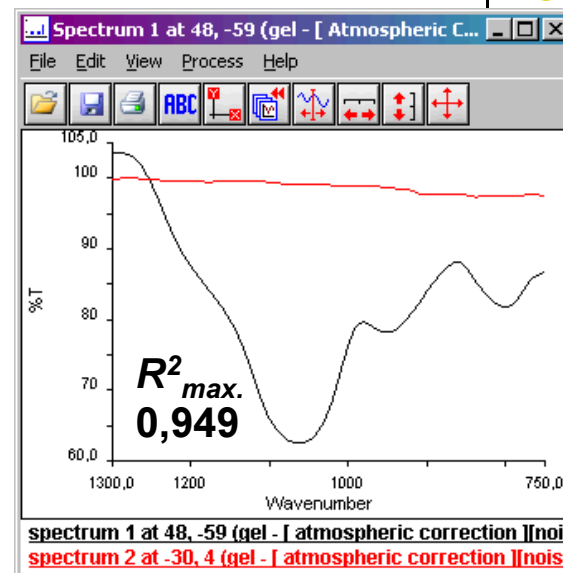
Technológiai probléma vs. imaging II.: változékony tartomány kiválasztása



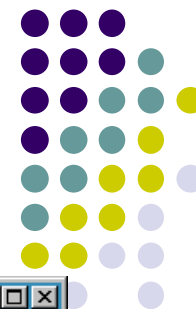
4000-750 cm⁻¹



1600-750 cm⁻¹

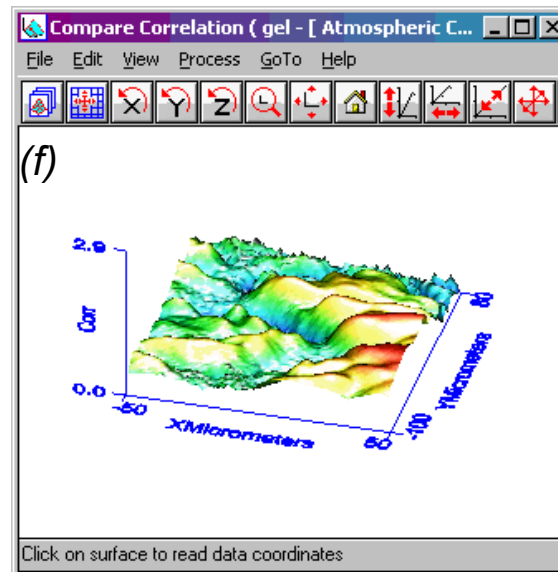
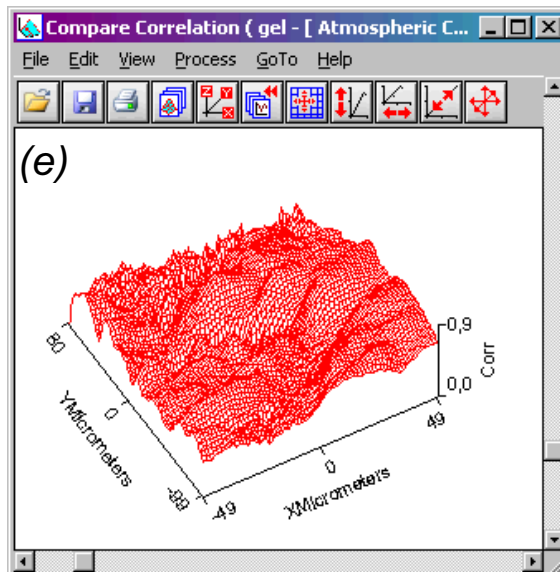
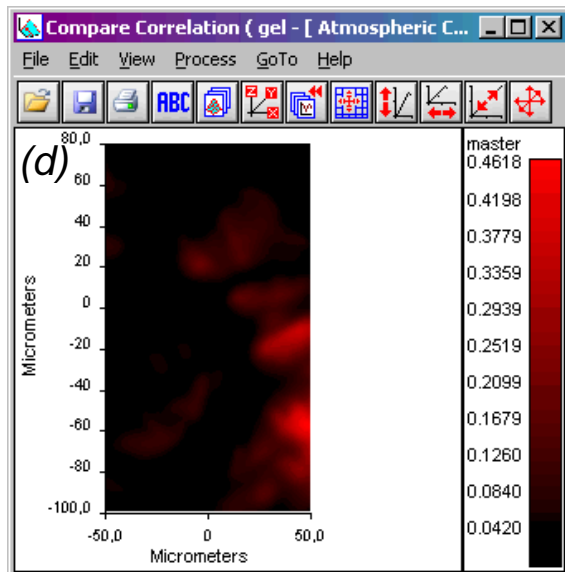
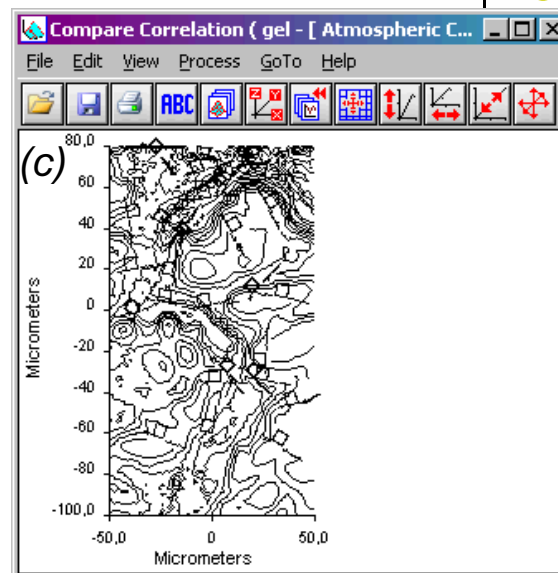
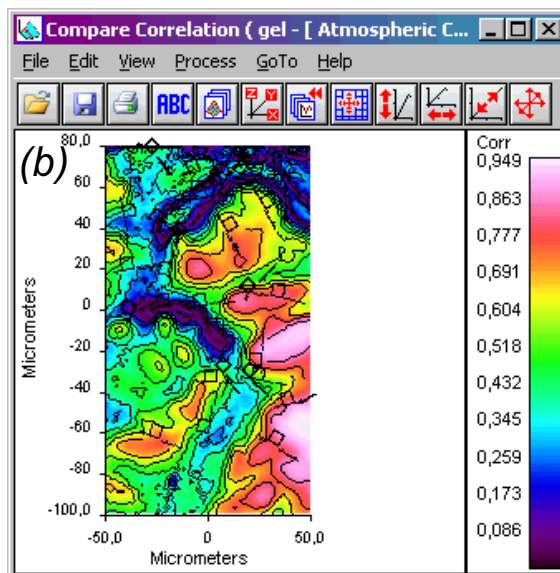
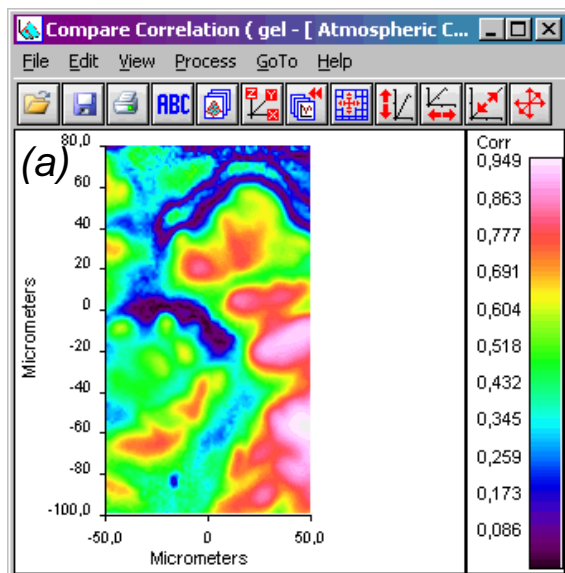


1300-750 cm⁻¹



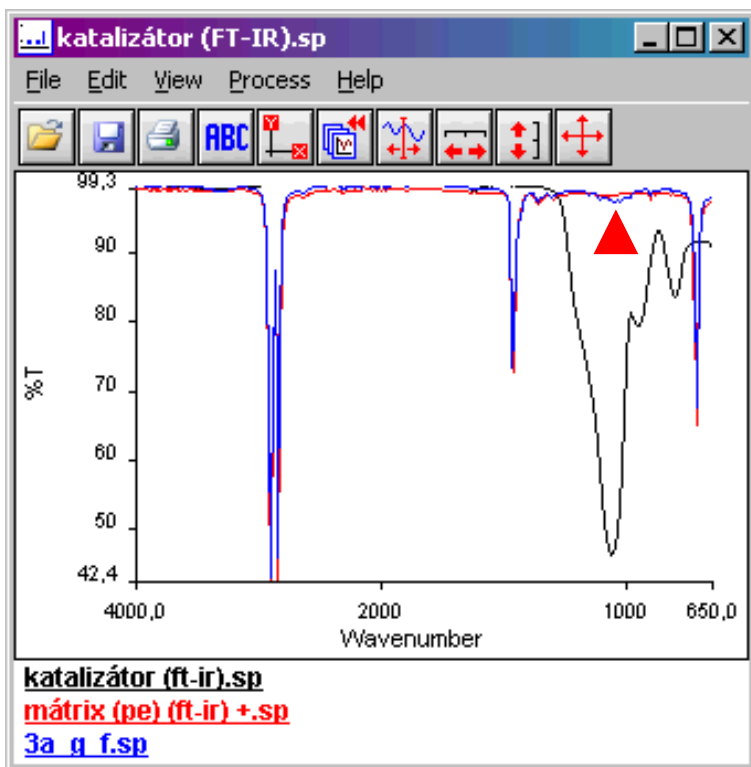
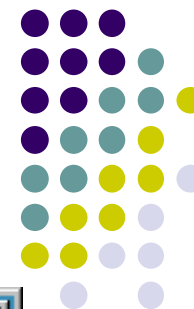
Technológiai probléma vs. imaging II.: megjelenítési lehetőségek

Gergely Szilveszter (BME)

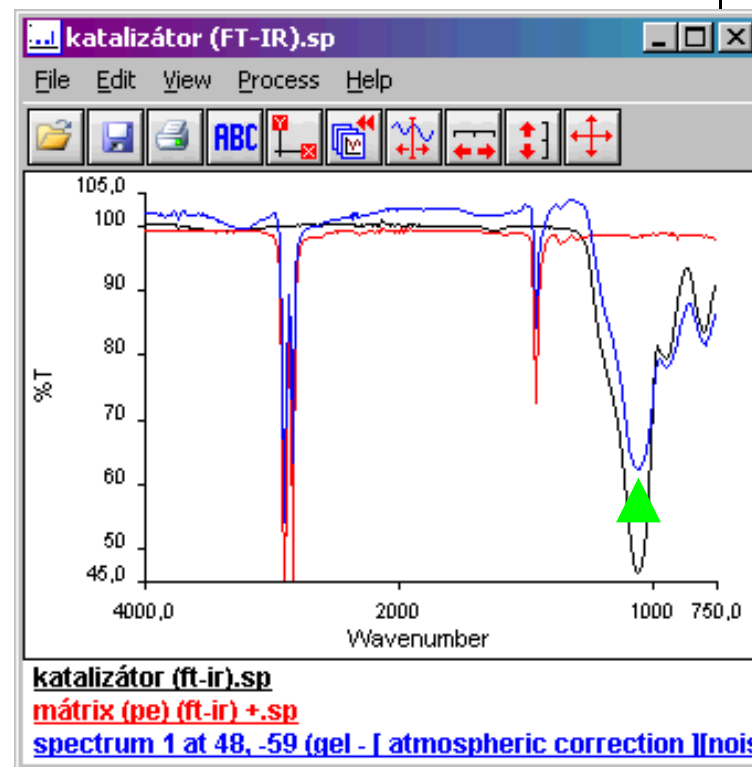


(a) False Color Image, (b) False Color Image with Contours, (c) Contour Map, (d) Overlay Image, (e) Wire Surface Projection, (f) 3D Surface Projection

Technológiai probléma vs. imaging II.: előnyök és hátrányok



- Makro ATR (UATR)
- gyors mérés (~percek)
- csak kvázi átlag sp. (~mm²)
- kis katalizátor jel

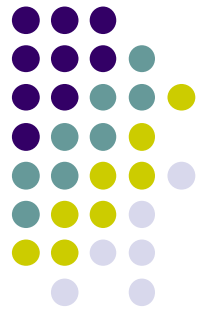


- Mikro ATR (imaging)
- lassabb mérés (~óra)
- képpontonkénti sp.-ok (~µm²)
- nagy katalizátor jel



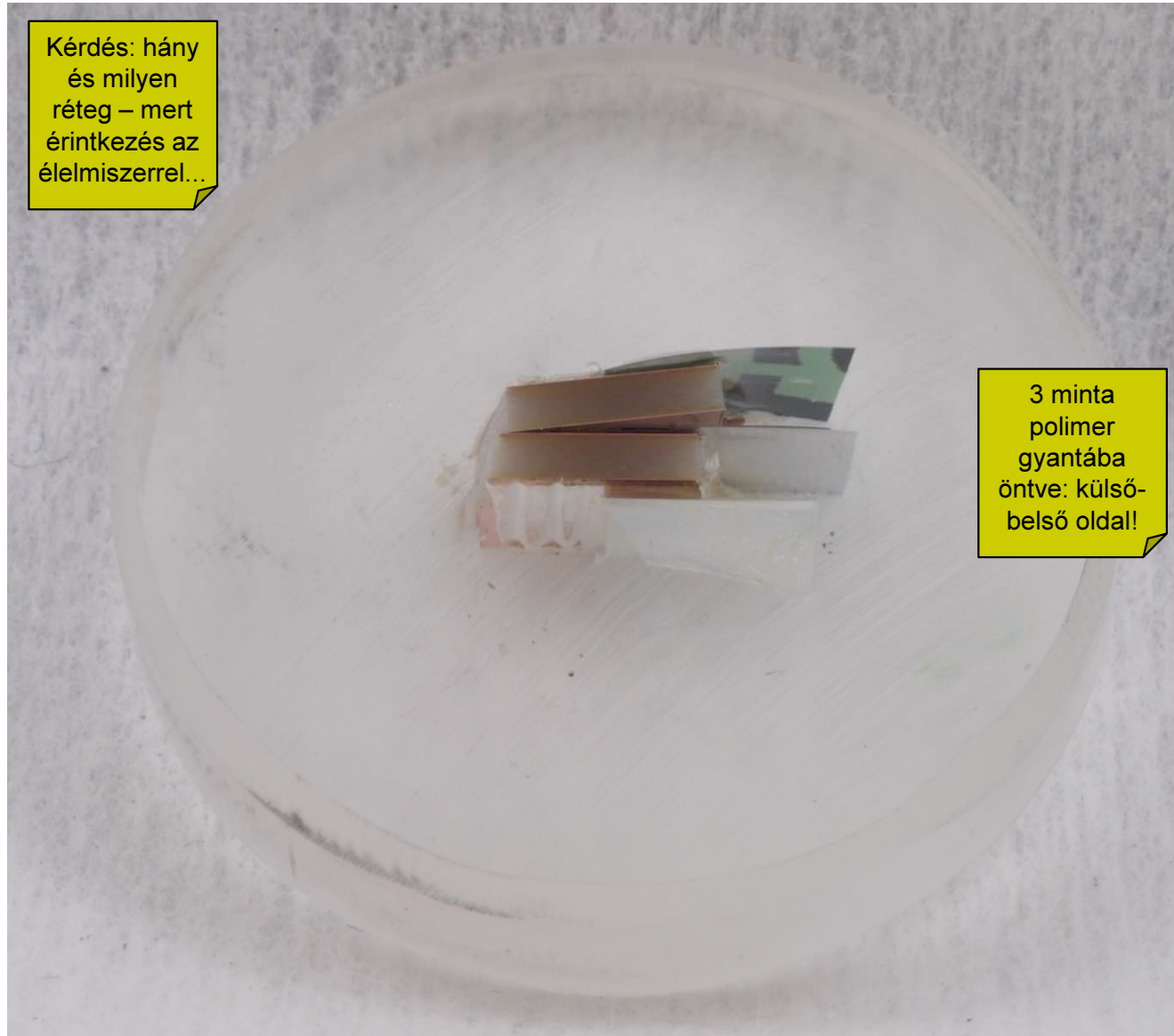
Csomagolóanyag – „as is”

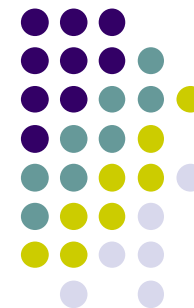
Lehetne
biológiailag
lebomló
csomagoló
anyag is, de...



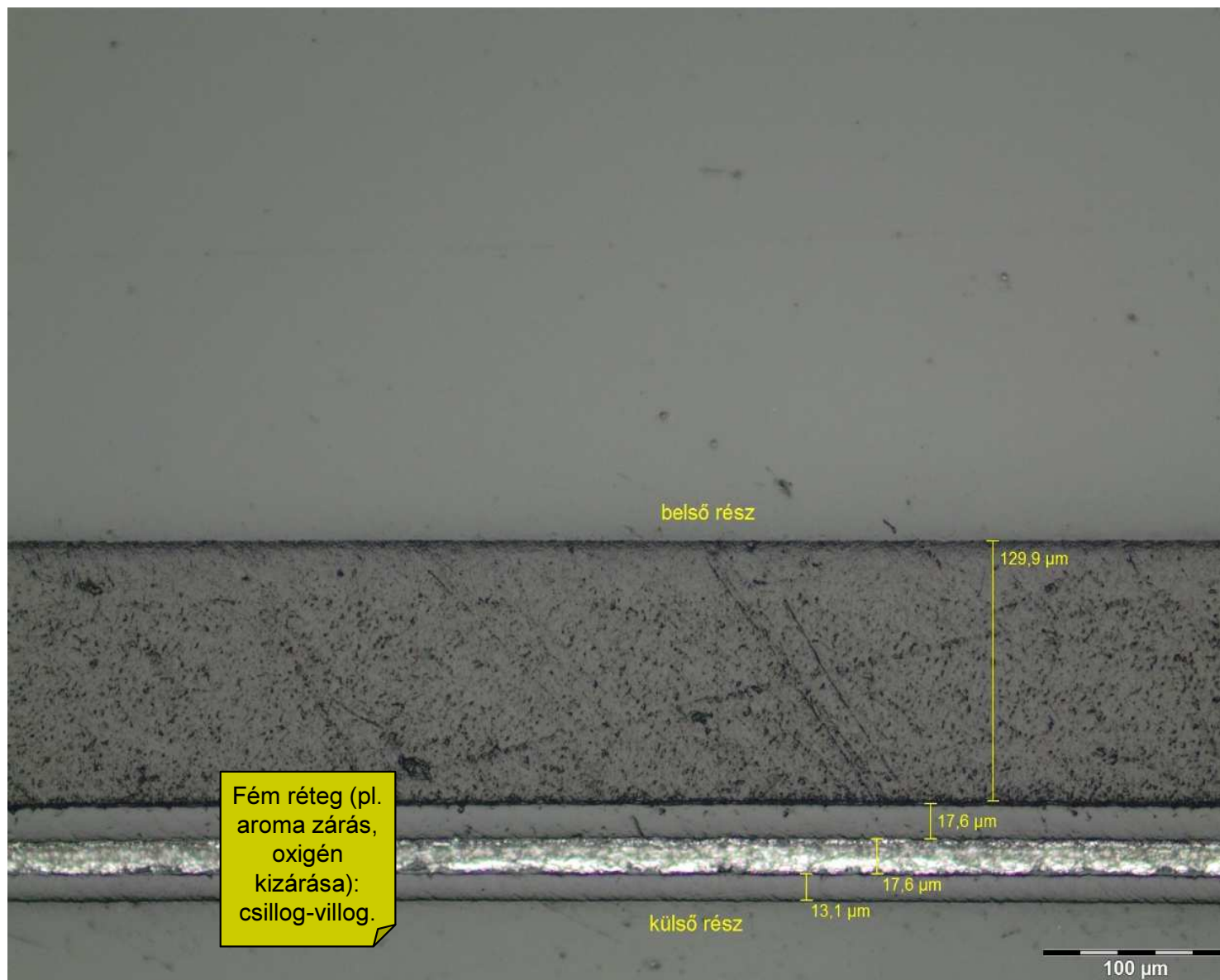
Kérdés: hány
és milyen
réteg – mert
érintkezés az
élelmiszerrel...

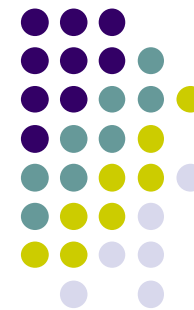
3 minta
polimer
gyantába
öntve: külső-
belső oldal!



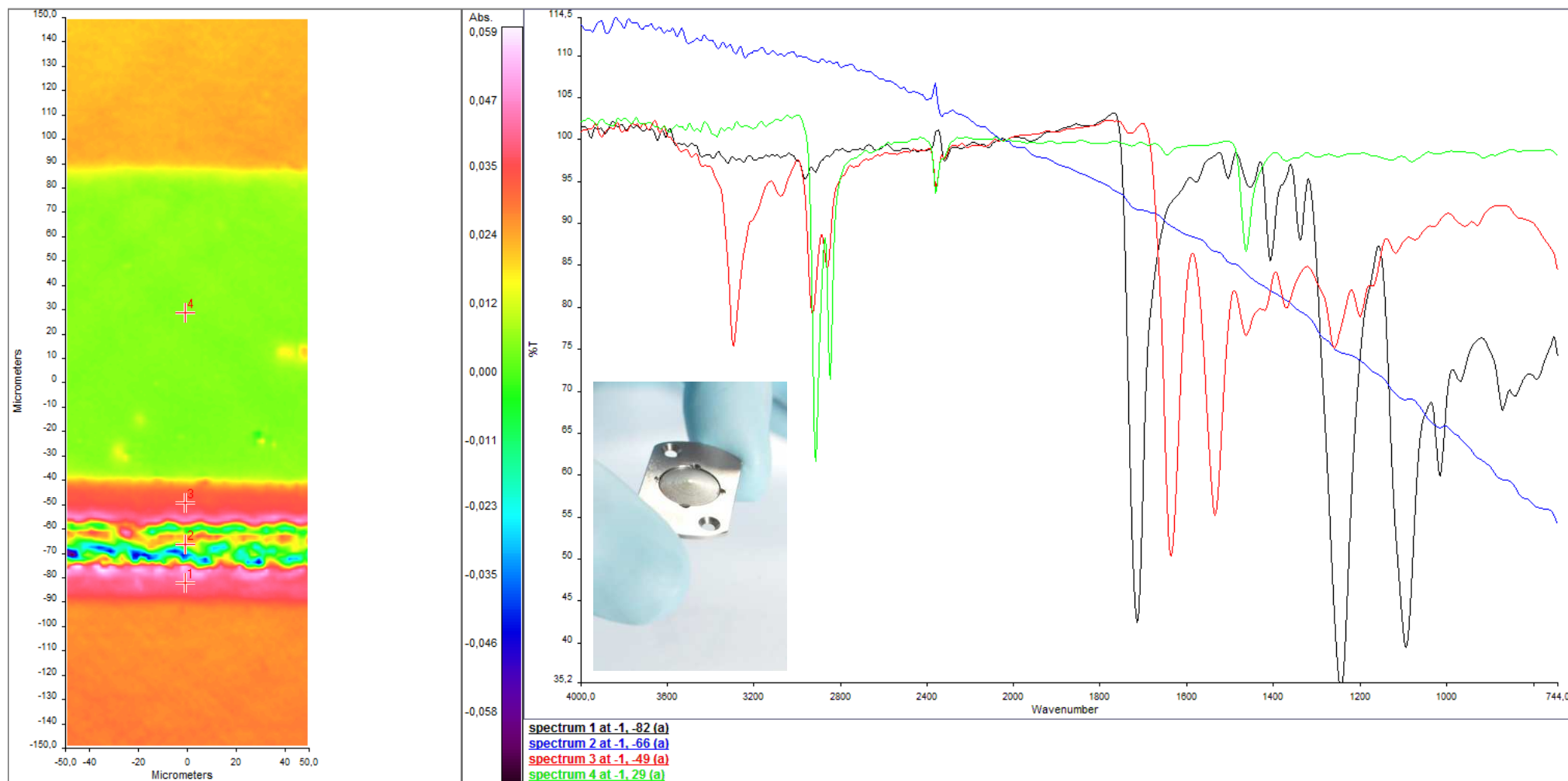


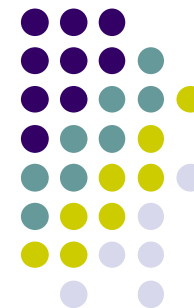
Csomagolóanyag – fénymikroszkóp



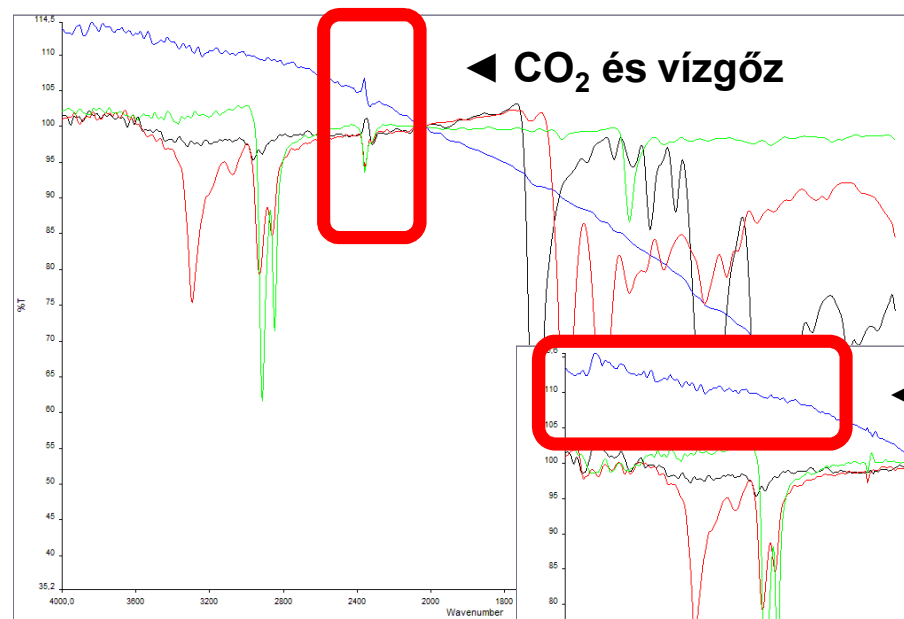


Csomagolóanyag – IR mikroszkóp

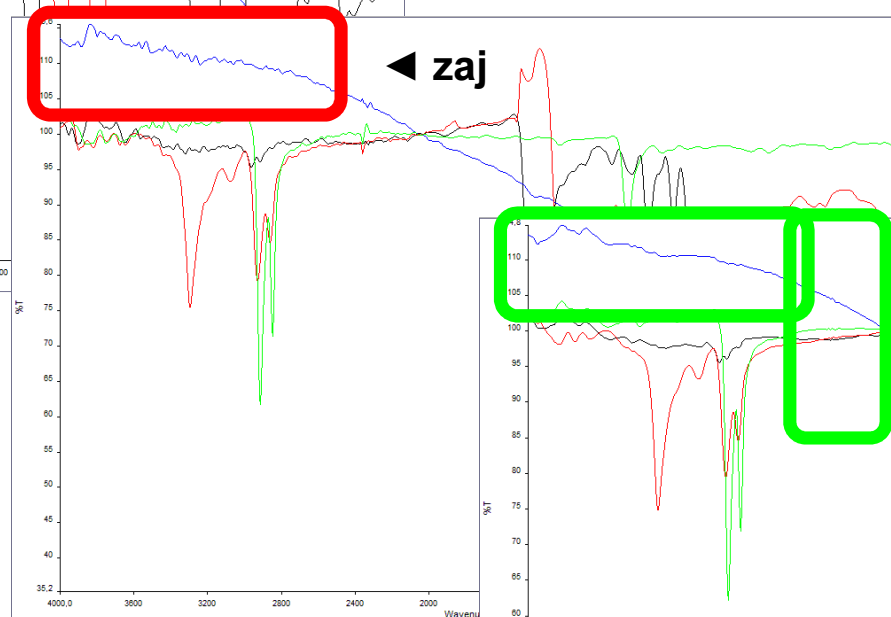




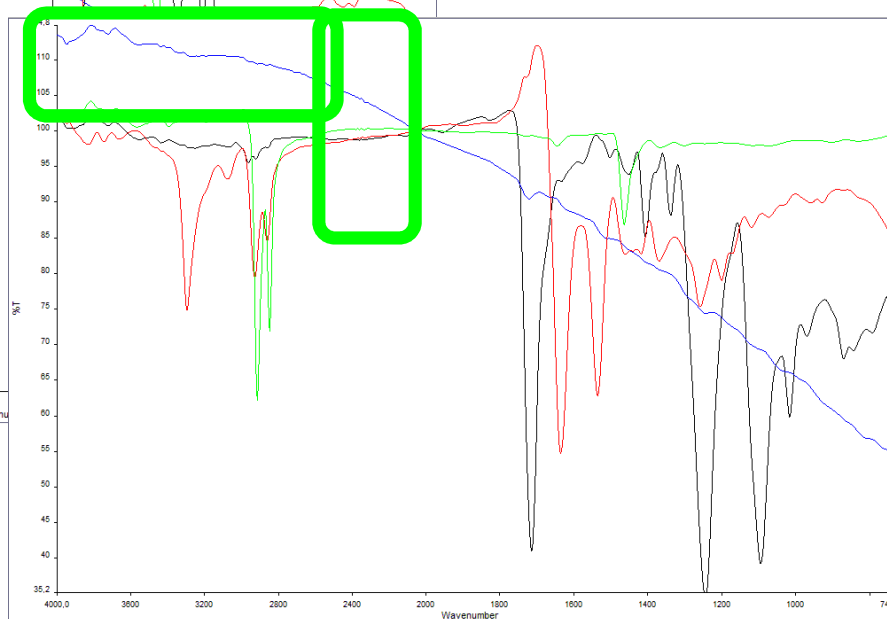
Csomagolóanyag – előkezelések



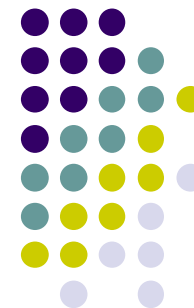
spectrum 1 at -1.32 (a)
spectrum 2 at -1.66 (a)
spectrum 3 at -1.49 (a)
spectrum 4 at -1.29 (a)



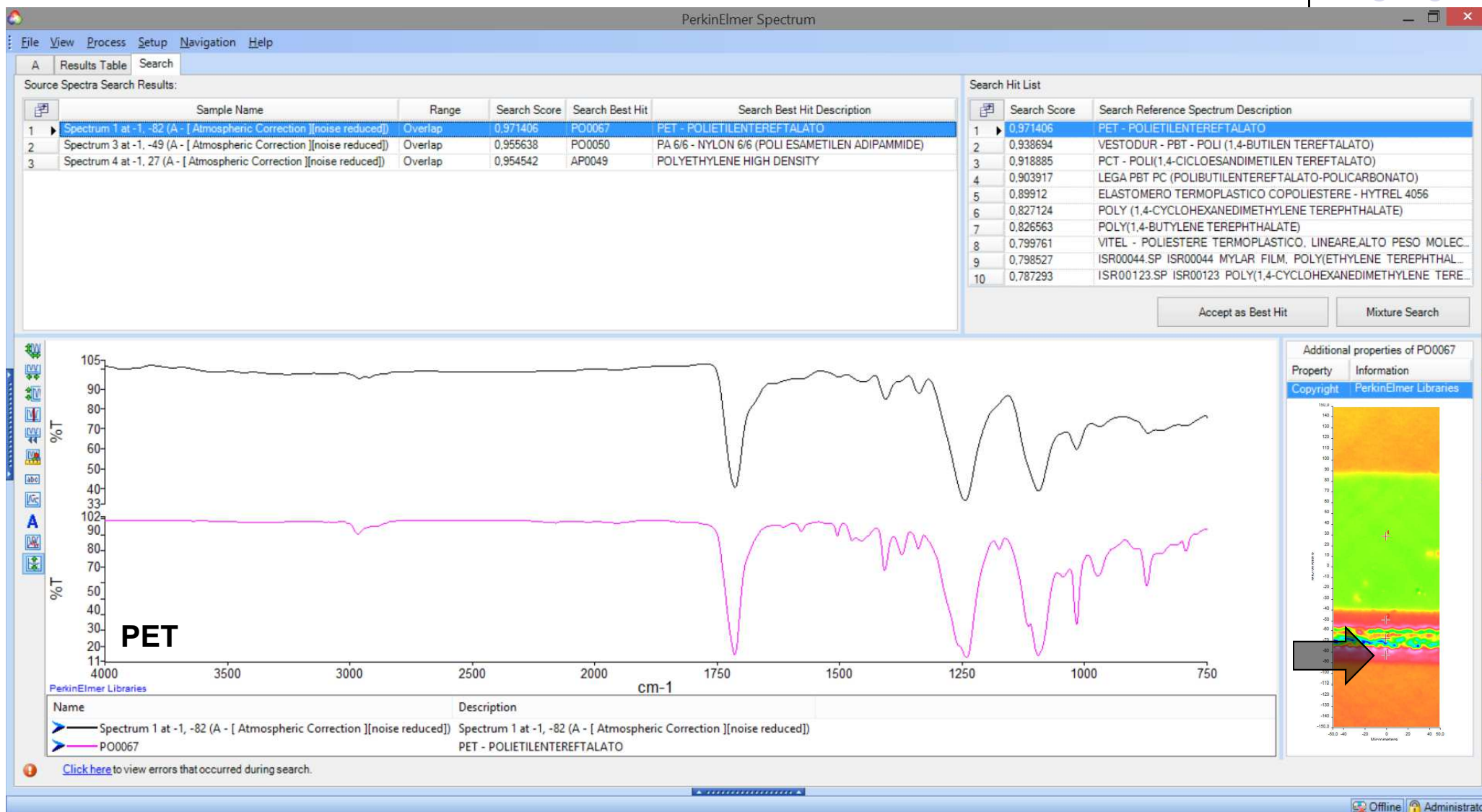
spectrum 1 at -1.32 (a - [atmospheric correction])
spectrum 2 at -1.66 (a - [atmospheric correction])
spectrum 3 at -1.49 (a - [atmospheric correction])
spectrum 4 at -1.29 (a - [atmospheric correction])

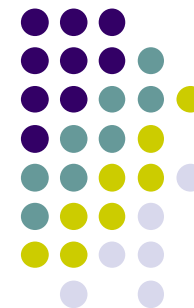


spectrum 1 at -1.32 (a - [atmospheric correction] [noise reduced])
spectrum 2 at -1.66 (a - [atmospheric correction] [noise reduced])
spectrum 3 at -1.49 (a - [atmospheric correction] [noise reduced])
spectrum 4 at -1.27 (a - [atmospheric correction] [noise reduced])

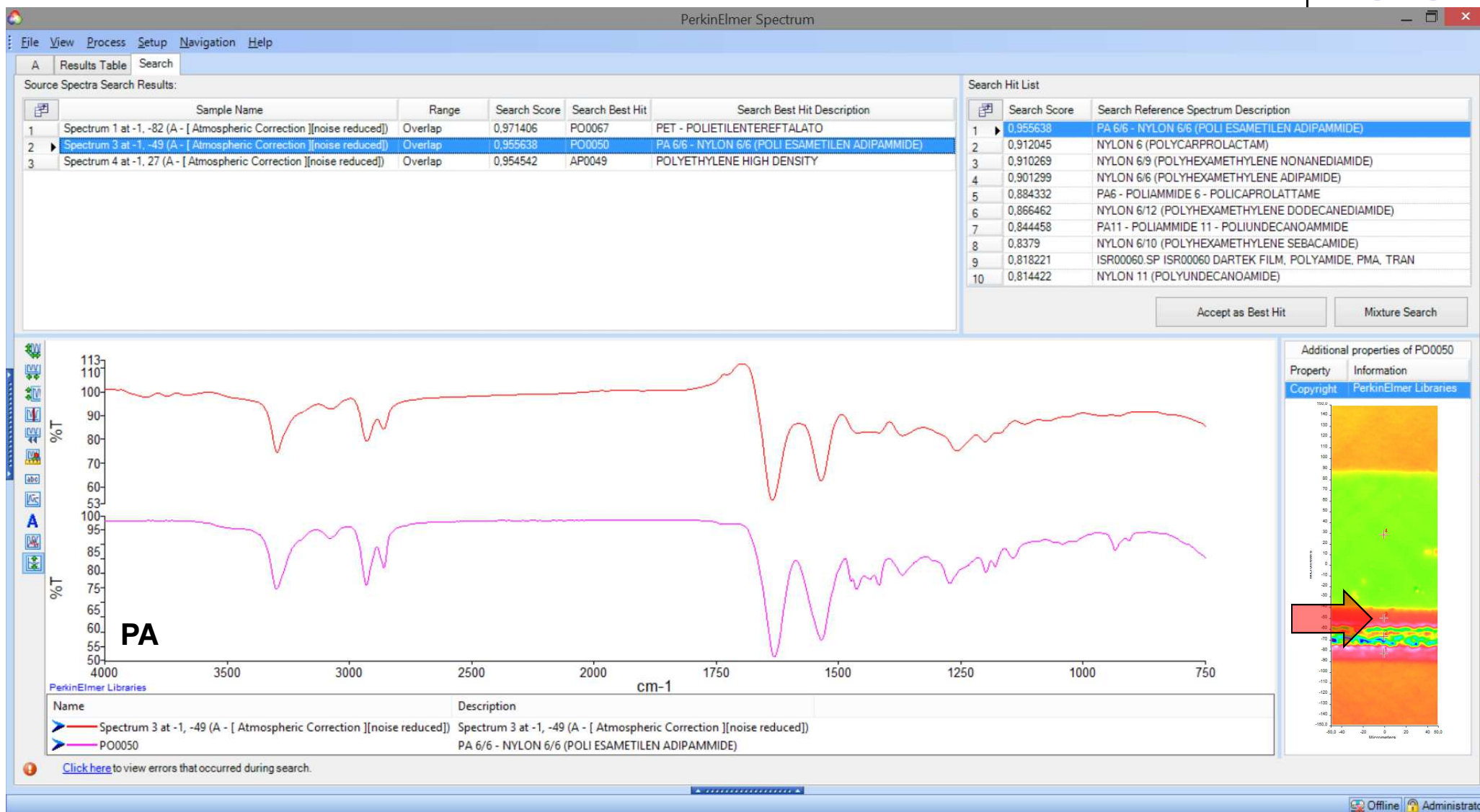


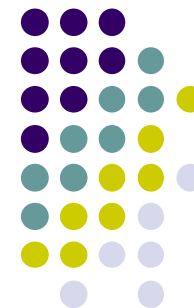
Csomagolóanyag – azonosítás I.



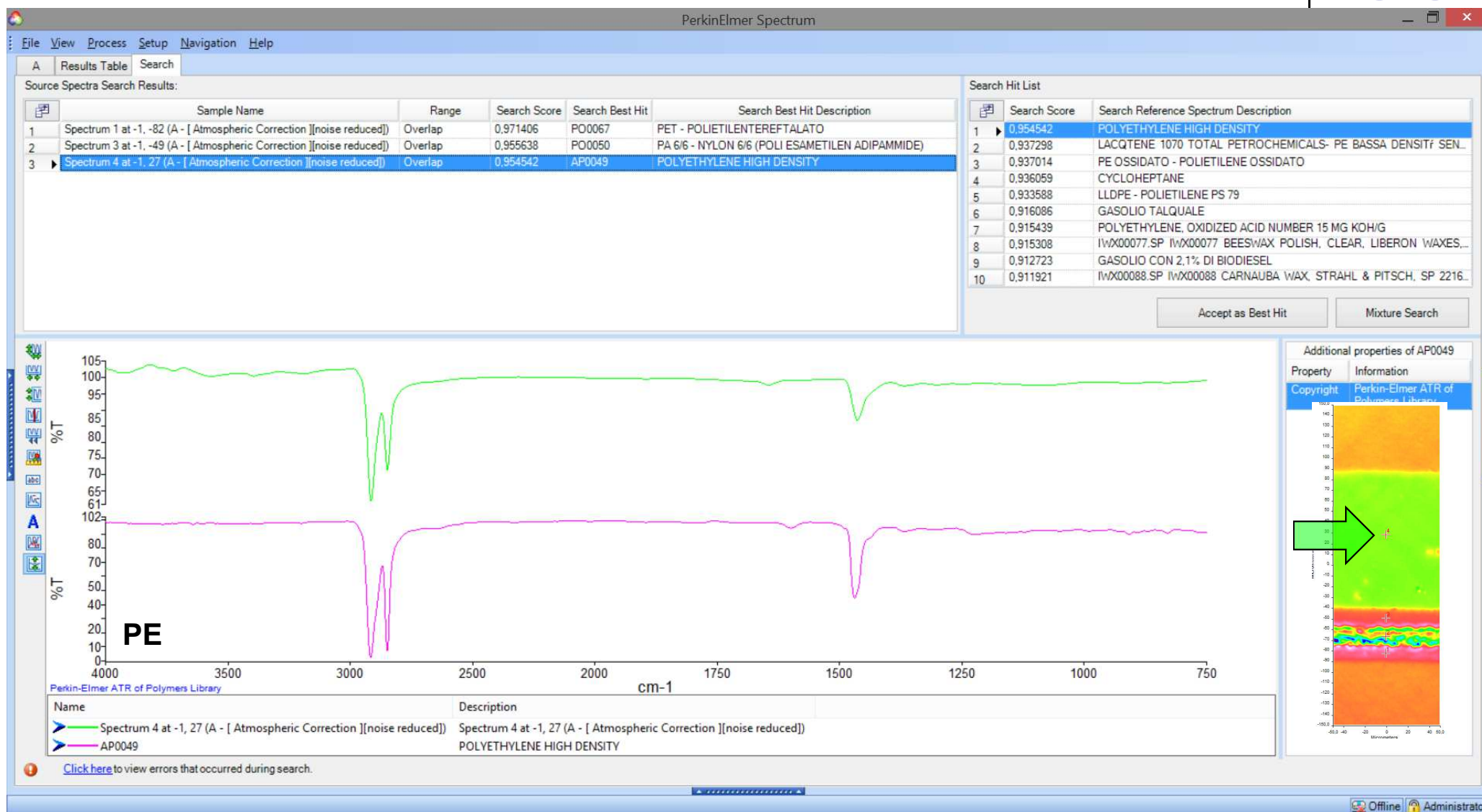


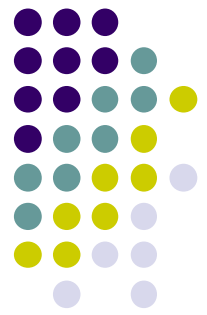
Csomagolóanyag – azonosítás II.



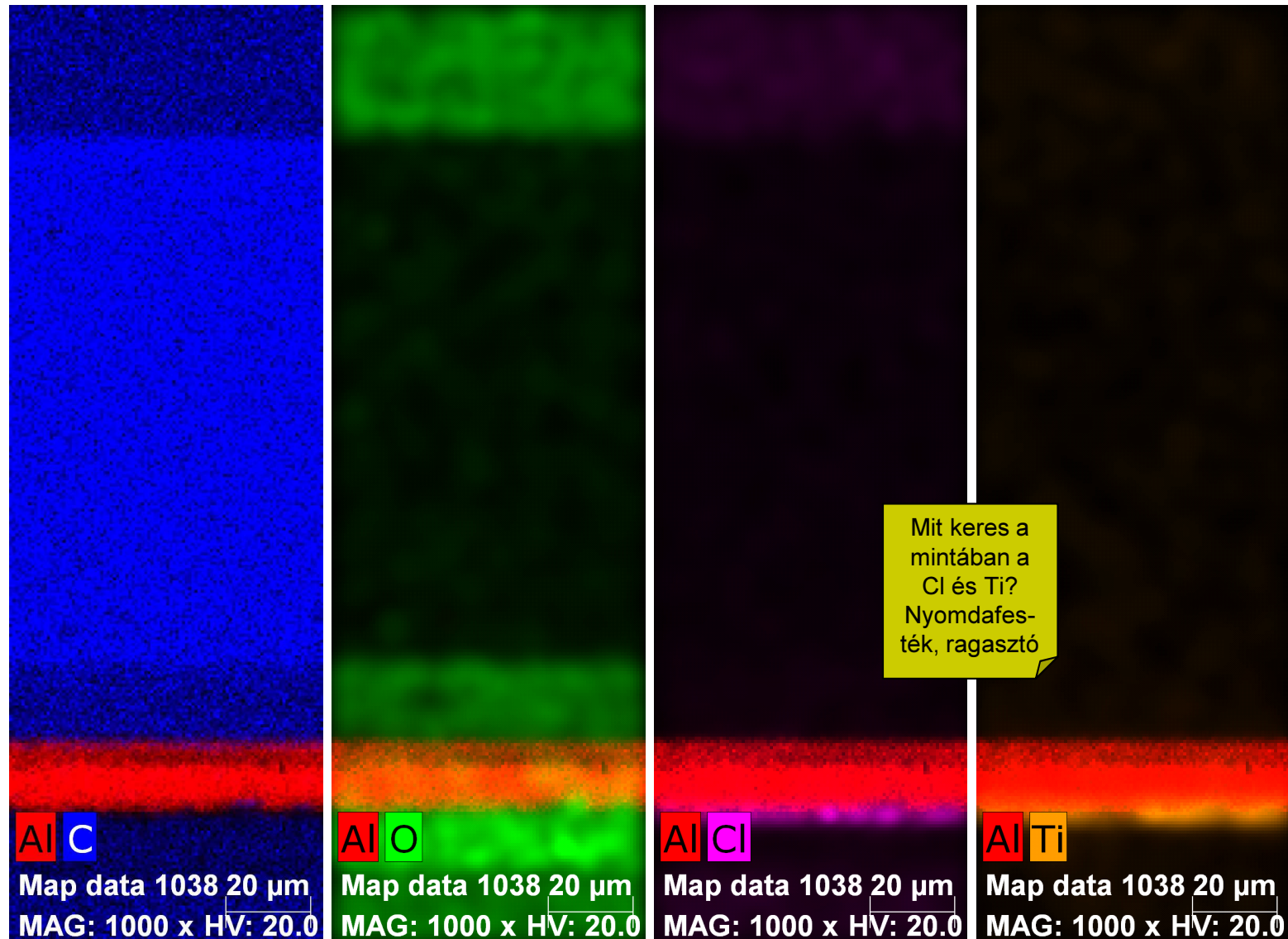


Csomagolóanyag – azonosítás III.

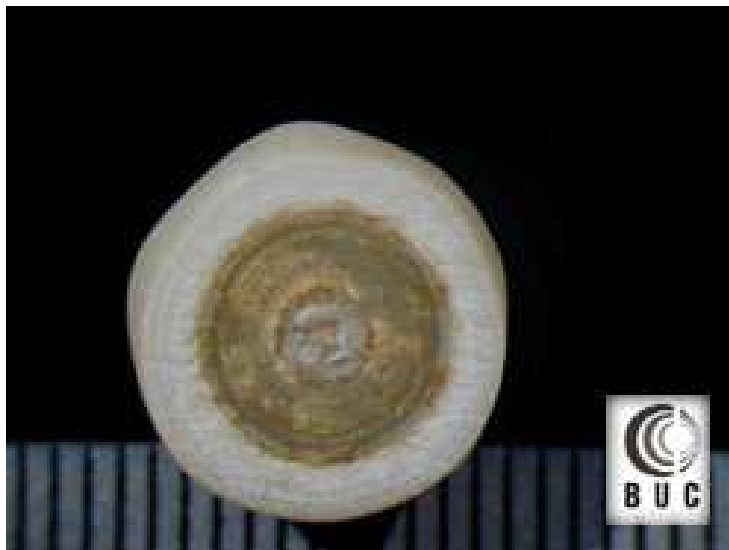
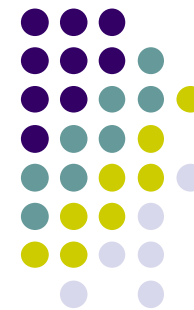




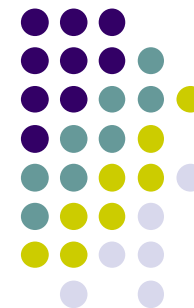
Csomagolóanyag – SEM



Altesti dolgaink – kő kövön



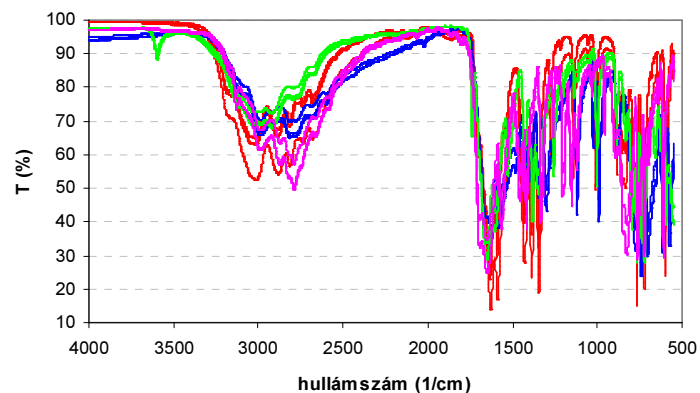
Kövek
(epe-, vese-,
húgykő)



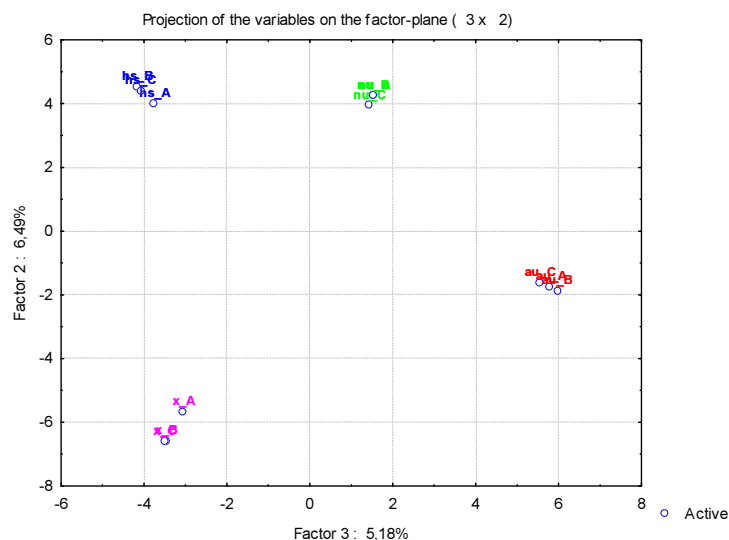
Altesti dolgaink – spektrumok és PCA

standardok

ammónium-hidrogén-urát (au), húgysav (hs),
nátrium-hidrogén-urát (nu), xantin (x)

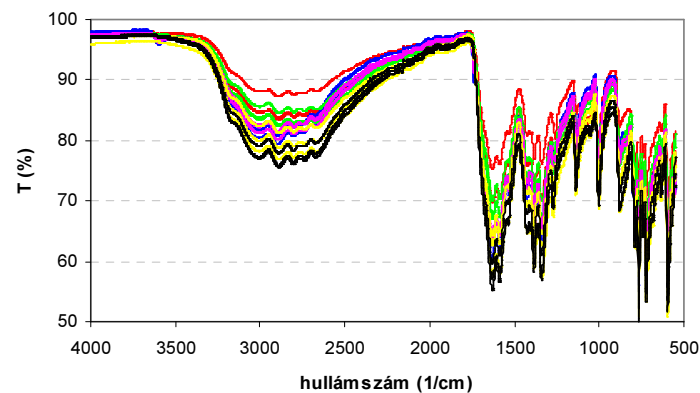


A kőalkotók
spektrumai,
eloszlása.

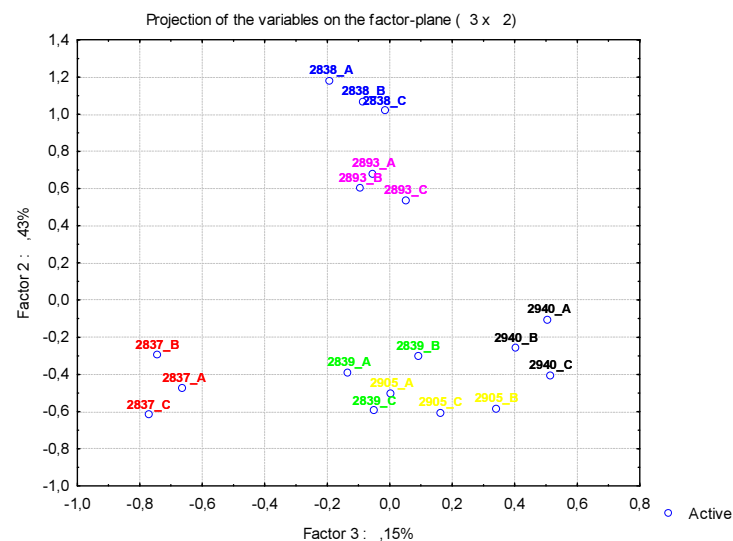


minták

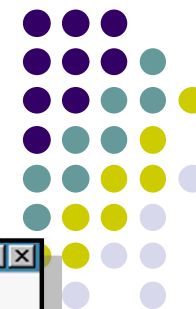
2837, 2838, 2839,
2893, 2905, 2940



A minták
spektrumai,
eloszlása.



Altesti dolgaink – keresés könyvtárral



Spectrum Express

File View Measurement Process Setup Navigation Help

Data Explorer minta | Results Table Compare

Source Spectra Compare Results:

Name(ID)	Best Hit	Correlation	Pass
1 2837_A	C:\nirs\BUC\...nu_B.sp	0,766407	Pass
2 2837_B	C:\nirs\BUC\...nu_B.sp	0,780973	Pass
3 2837_C	C:\nirs\BUC\...nu_B.sp	0,763029	Pass
4 2838_A	C:\nirs\BUC\...nu_B.sp	0,856915	Pass
5 2838_B	C:\nirs\BUC\...nu_B.sp	0,840922	Pass
6 2838_C	C:\nirs\BUC\...nu_B.sp	0,834263	Pass
7 2839_A	C:\nirs\BUC\...au_B.sp	0,671768	Pass
8 2839_B	C:\nirs\BUC\...au_B.sp	0,675275	Pass
9 2839_C	C:\nirs\BUC\...au_B.sp	0,6536	Pass
10 2893_A	C:\nirs\BUC\...nu_B.sp	0,791473	Pass
11 2893_B	C:\nirs\BUC\...nu_B.sp	0,795315	Pass
12 2893_C	C:\nirs\BUC\...nu_B.sp	0,791055	Pass
13 2905_A	C:\nirs\BUC\...au_B.sp	0,692841	Pass
14 2905_B	C:\nirs\BUC\...au_B.sp	0,675795	Pass

Compared References:

Name(ID)	Correlation	Factor	Correlation Criteria	Pass / Fail
1 C:\nirs\BUC\...nu_B.sp	0,766407	0,167222	0,6	Pass
2 C:\nirs\BUC\...nu_A.sp	0,765956	0,166184	0,6	Pass
3 C:\nirs\BUC\...nu_C.sp	0,761803	0,188117	0,6	Pass
4 C:\nirs\BUC\...au_B.sp	0,619801	0,0799834	0,6	Pass
5 C:\nirs\BUC\...au_A.sp	0,541383	0,0919562	0,6	Fail
6 C:\nirs\BUC\...au_C.sp	0,536935	0,0963874	0,6	Fail
7 C:\nirs\BUC\...x_C.sp	0,137009	0,0227703	0,6	Fail
8 C:\nirs\BUC\...st...x_A.sp	0,136853	0,0289476	0,6	Fail
9 C:\nirs\BUC\...st...x_B.sp	0,135715	0,0226842	0,6	Fail
10 C:\nirs\BUC\...hs_A.sp	0,00131197	-0,00033...	0,6	Fail
11 C:\nirs\BUC\...hs_C.sp	0,0009337...	-0,00019...	0,6	Fail
12 C:\nirs\BUC\...hs_B.sp	0,00030344	-6,39077...	0,6	Fail

%T

cm-1

Name **Description**

2837_A
nu_B.sp

Setup Compare Options

Path	Correlation	Include
C:\nirs\BUC\standard\au	0,60	☒
C:\nirs\BUC\standard\hs	0,60	☒
C:\nirs\BUC\standard\nu	0,60	☒
C:\nirs\BUC\standard\x	0,60	☒

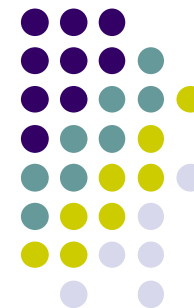
Setup Compare References

Add
Remove
Clear

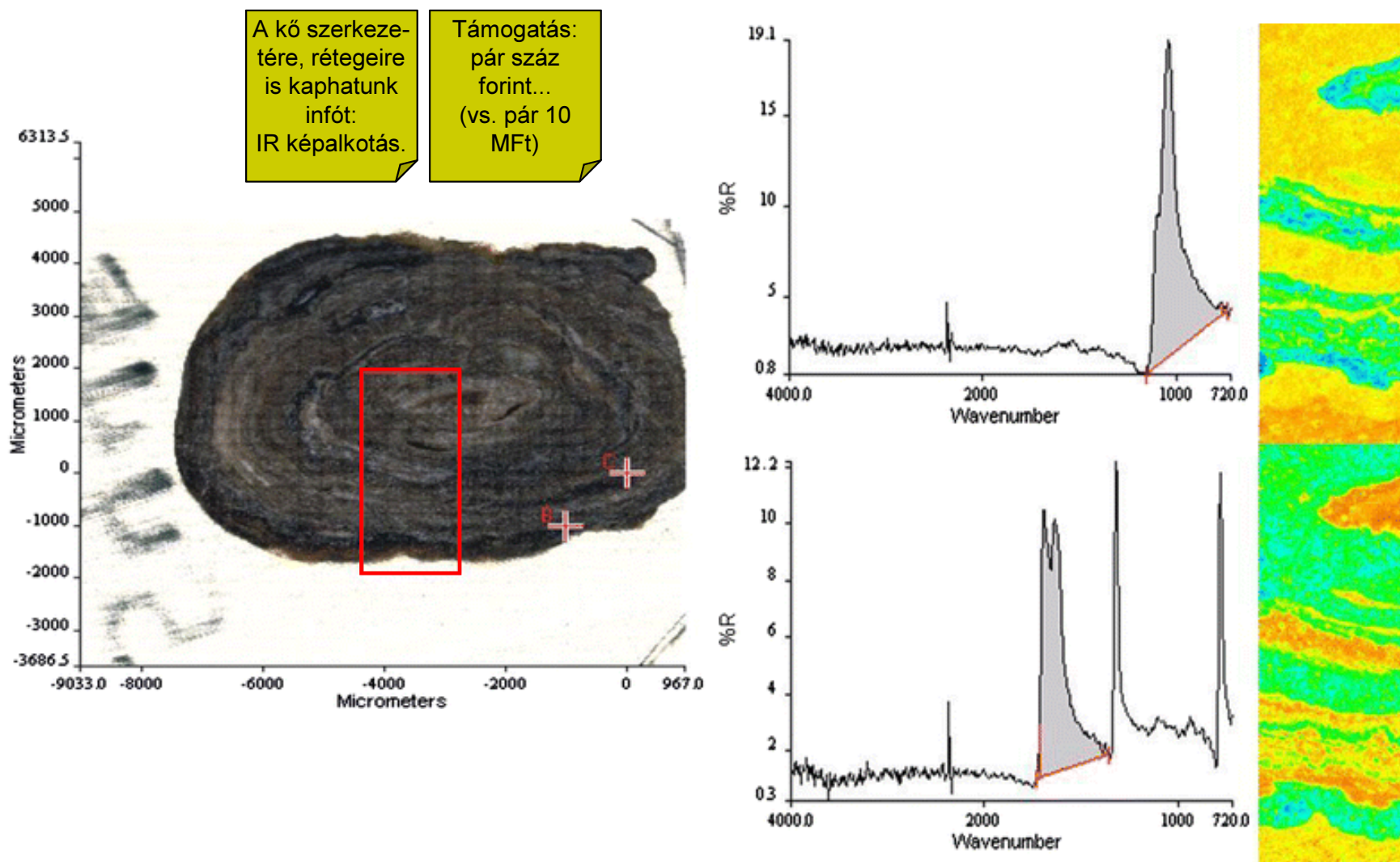
Pass / Fail
☒ Correlation
☐ Discrimination

Offline Administrator

Keresés spektrum-azonosság alapján.



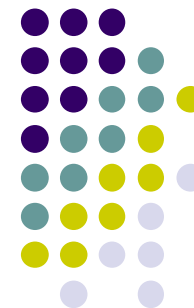
Altesti dolgaink – mire jó a kép? I.



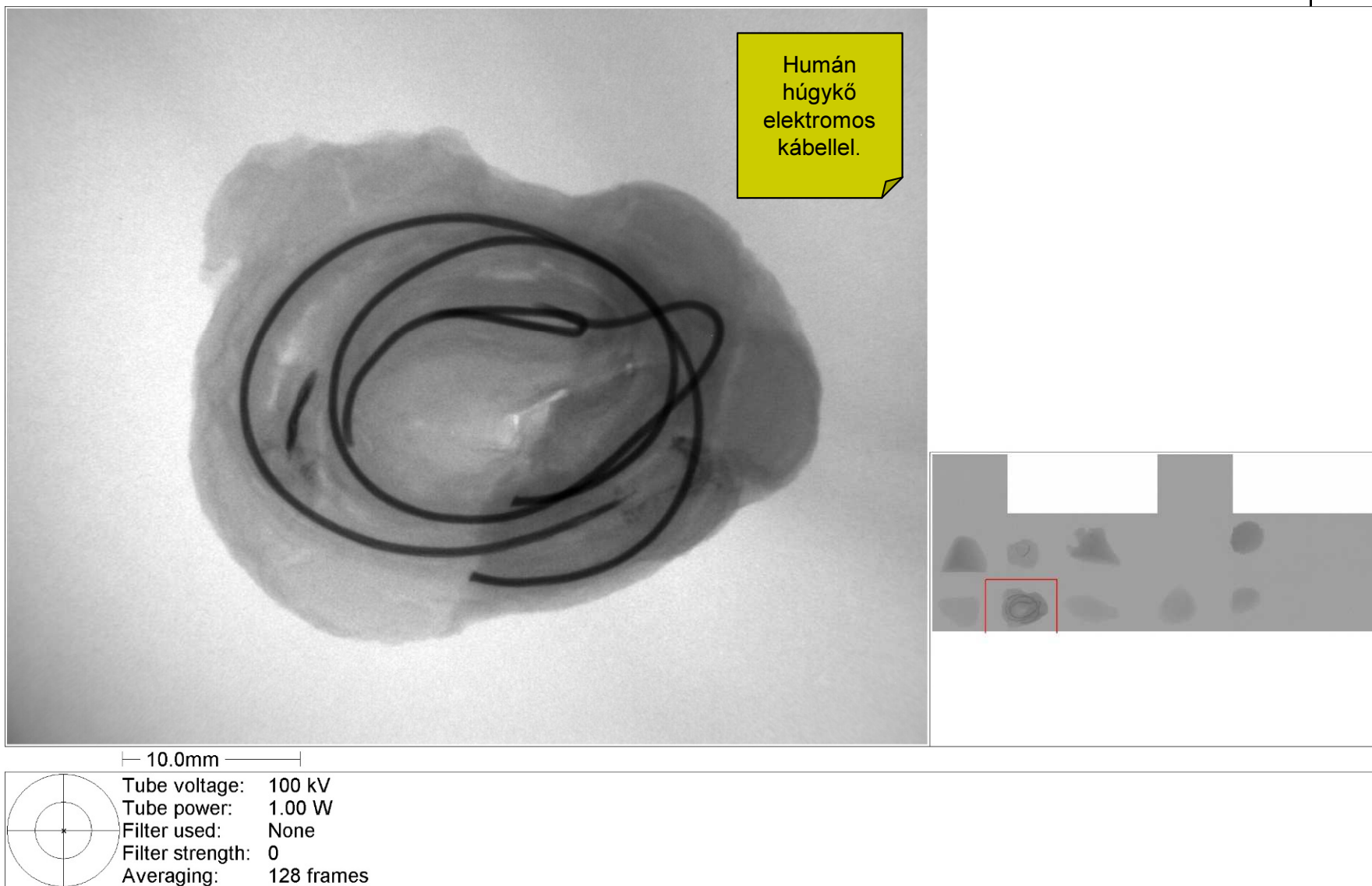
Anderson J.C. *et al.*:

Analysis of urinary calculi using an infrared microspectroscopic surface reflectance imaging technique.

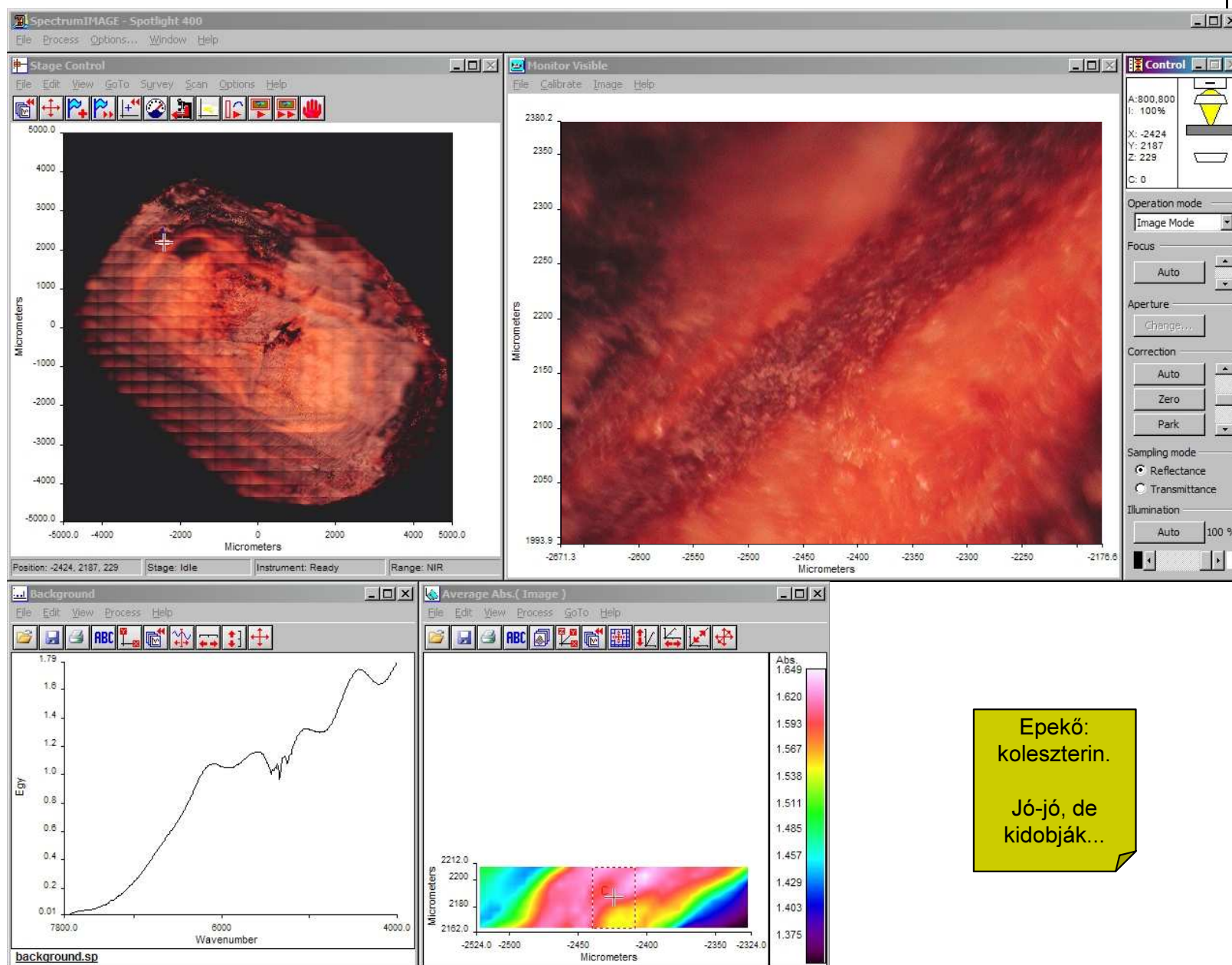
Urol. Res. (2007) 35(1) 41–48 (2007)



Altesti dolgaink – mire jó a kép? +



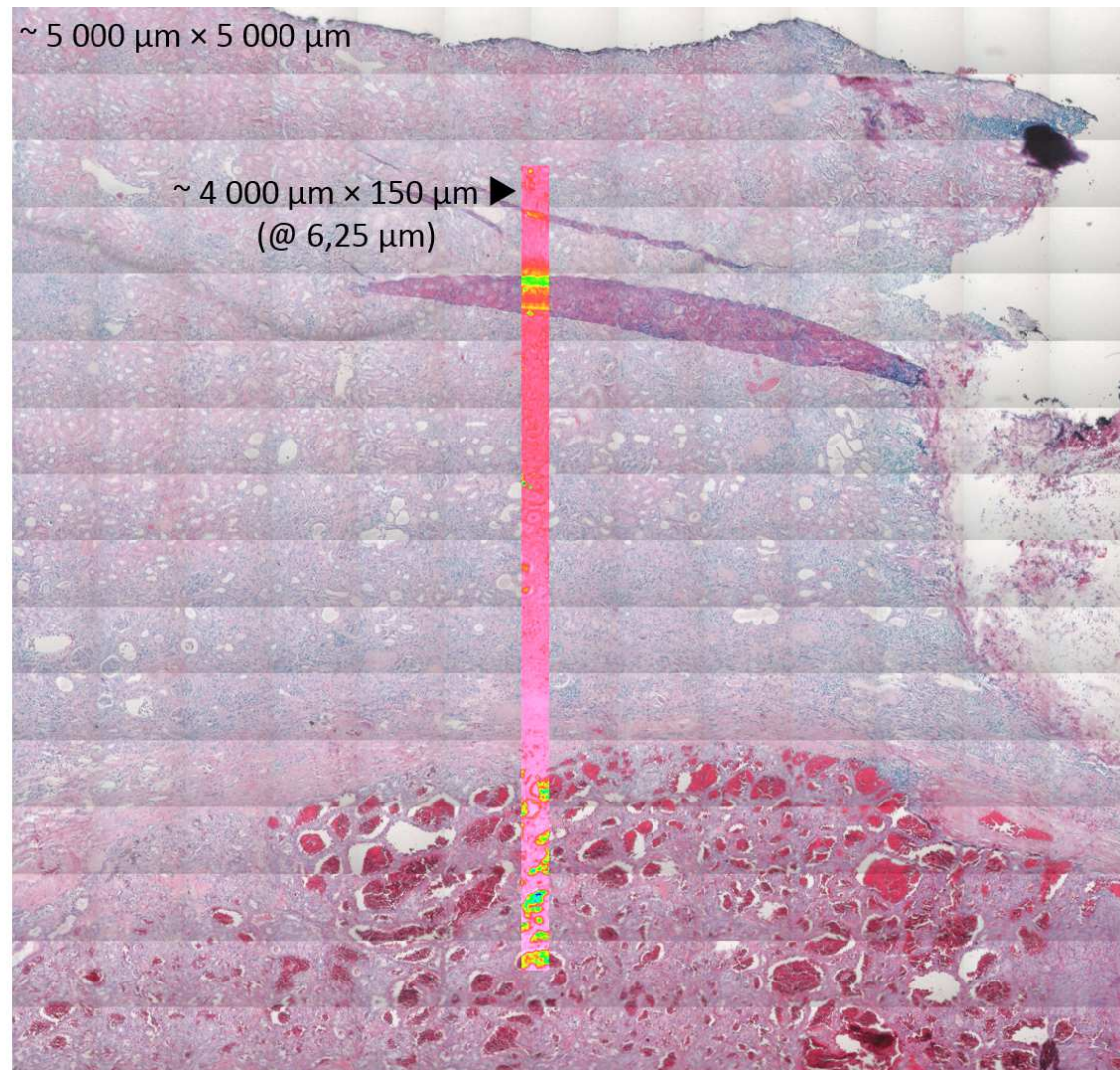
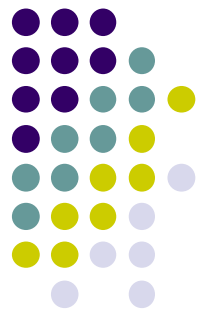
Altesti dolgaink – mire jó a kép? II.



Epekő:
koleszterin.

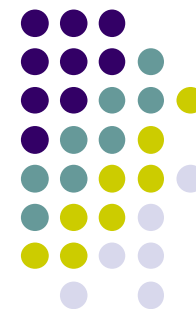
Jó-jó, de
kidobják...

Altesti dolgaink – mire jó a kép? III.

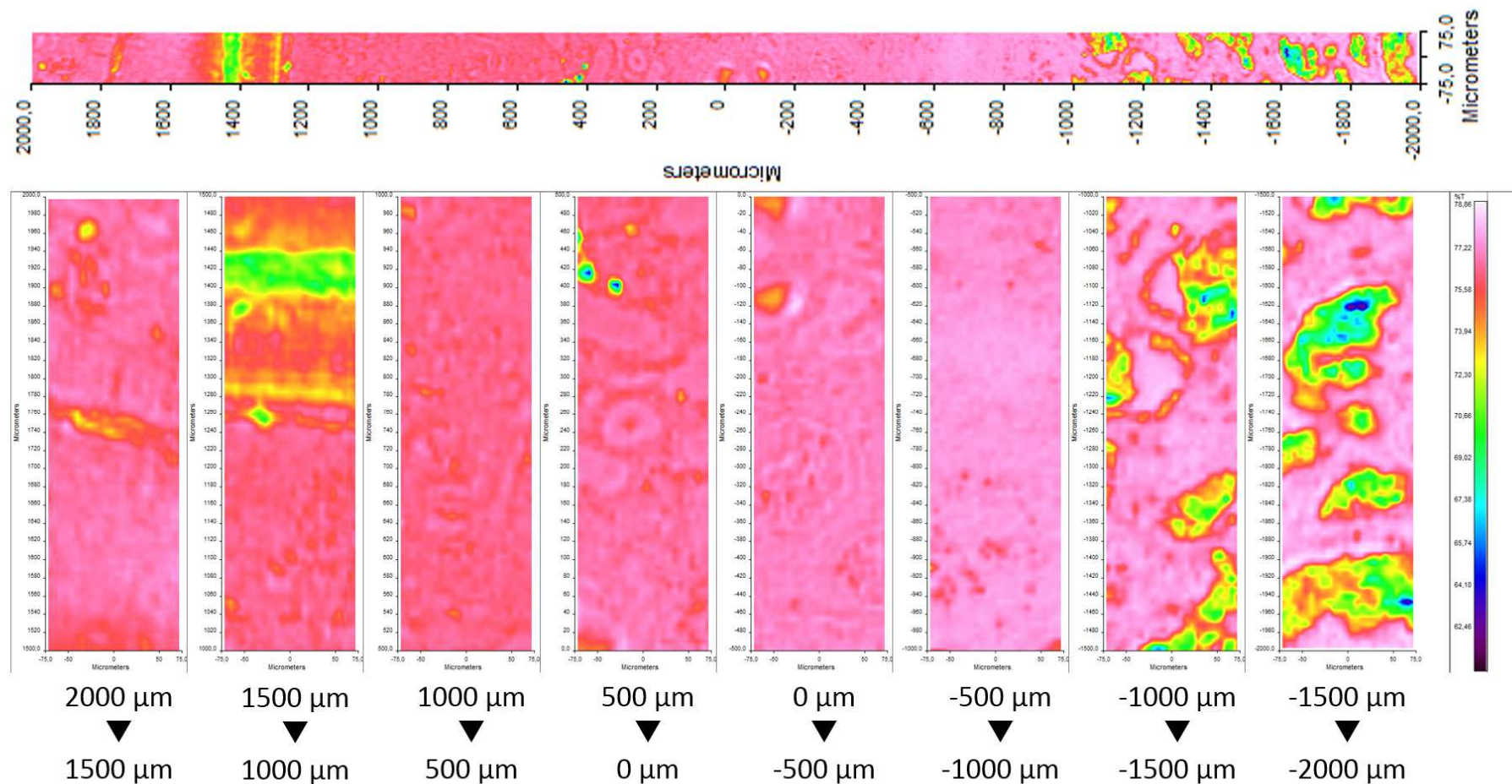


Mi jó, ha
kidobják?

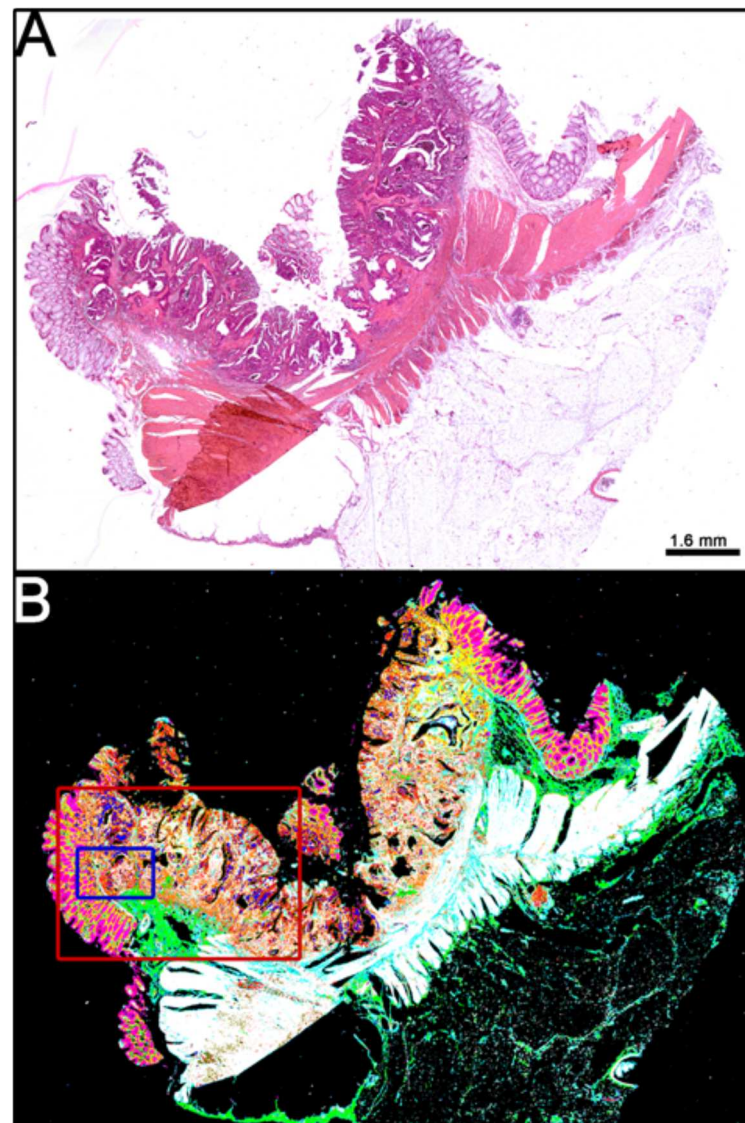
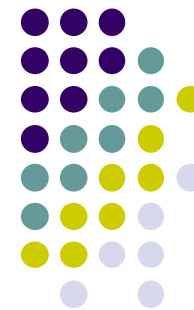
A daganat.



Altesti dolgaink – mire jó a kép? IV.



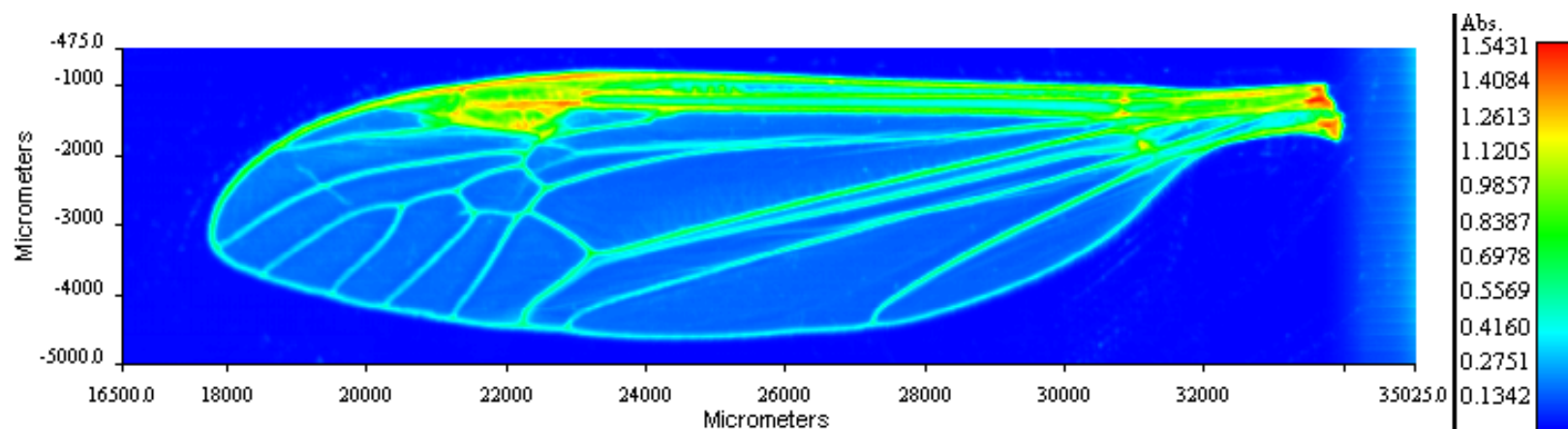
Altesti dolgaink – mire jó a kép? V.



A. Kallenbach-Thieltges *et al.*:
Immunohistochemistry, histopathology and infrared spectral histopathology of colon cancer tissue sections
J. Biophotonics 6(1) 88–100 (2013)

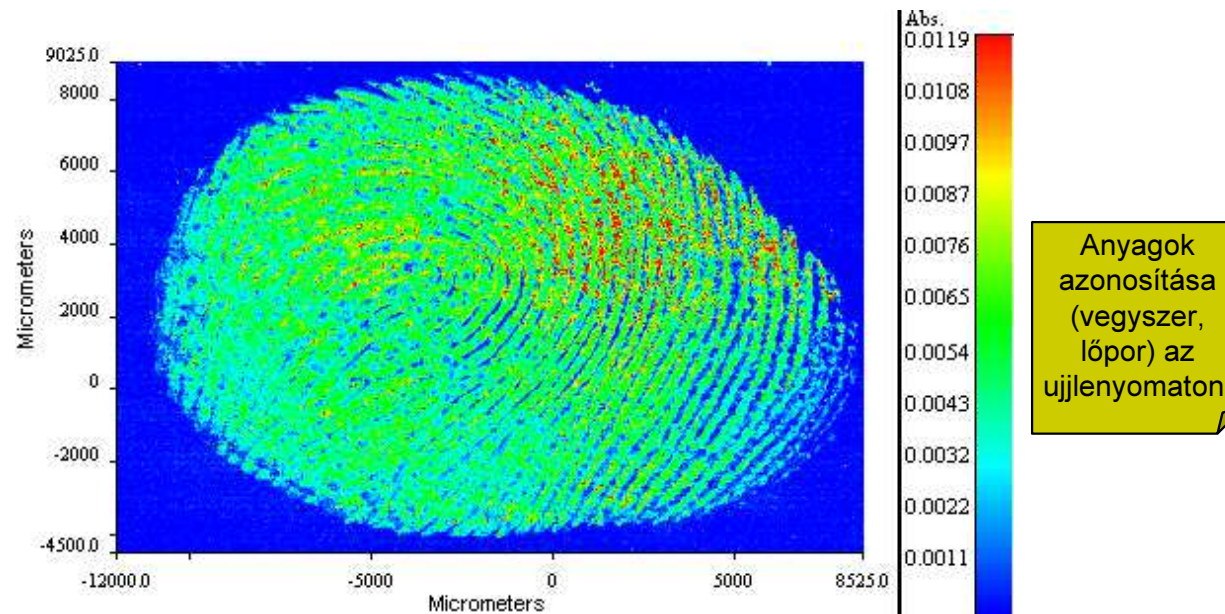


Képes képtelenségek



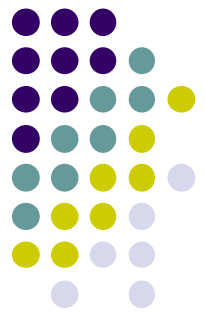
Szerkezet,
összetétel
megállapítása
roncsolás
nélkül.

lőszúnyog szárnya: 18,5 mm × 4,5 mm = 132 000 spektrum



Anyagok
azonosítása
(vegyszer,
lőpor) az
ujjlenyomaton.

ujjlenyomat: 20 mm × 13,5 mm = 450 000 spektrum



A (N)IR képalkotásról összefoglalva

- Roncsolásmentes (sok esetben, ha nem...)
- Relatív gyors (jelsorozatok ma már órás helyett perces nagyságrendben képezhetők)
- Rengeteg rejtett információ megfejthető (adatbányászat, oknyomozások)
- Visszacsatolás a technológiába, minőségellenőrzésbe, minőségbiztosításba
- Tudásintenzív módszerek
- Nem olcsó (de nem az LN2 miatt drága)

Köszönöm megtisztelő figyelmüket!

Gergely Szilveszter

Budapesti Műszaki és Gazdaságtudományi Egyetem

Alkalmazott Biotechnológia és Élelmiszertudományi Tanszék



M Ű E G Y E T E M 1 7 8 2

PerkinElmer Szeminárium

2015. október 20.

black box



grey box

