# MOLECULAR IMAGING

## Functional Imaging Methods
## Multi-Modality

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# Overview

- What do we call molecular imaging and why?
- What can we exploit to obtain molecular imaging data?
- Research-oriented molecular imaging methodologies (in general)
- Clinical application possibilities of molecular imaging
  (screening, diagnostics, personalized therapy, monitoring/follow-up)
- Most actually important methods in M.I., outlook for tomorrow

  PET, SPECT, MRI, Planar Fluorescence, Optical Tomographies)

  Onkológia, idegtudomány, kardiovaszkuláris medicina, reumatológia, endokrinológia, sebészet
- Correlation of functional and morphological information
- PACS and clinical relevance of image segmentation/registration

- Molecular Biology + In-Vivo Non-Invasive Imaging?

  “Imaging is the extraction of information in time and space at all levels of biological organization"

  (Dr. Elias Zerhouni, XIVth NIH Director)

“Molecular imaging is the visualization, characterization, and measurement of biological processes at the molecular and cellular levels in humans and other living systems.”

(definition by the U.S. Society of Nuclear Medicine and Molecular Imaging)

# Molecular Imaging Ideals:

Biological

Visualization ⟷ Characterization ⟷ Measurement

Signal to Noise Ratio

Accuracy of Information (where?) Spatial Resolution (yes or no?)

Time Resolution (when?) (how?)

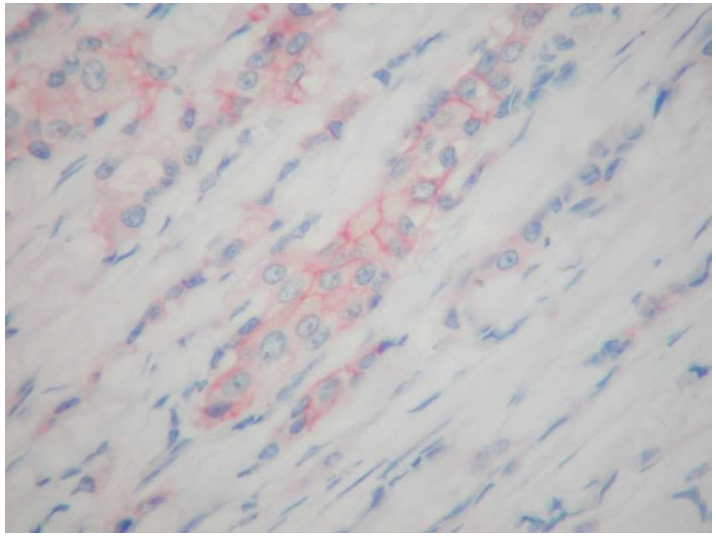
Magnitude and Density of Information (how?)

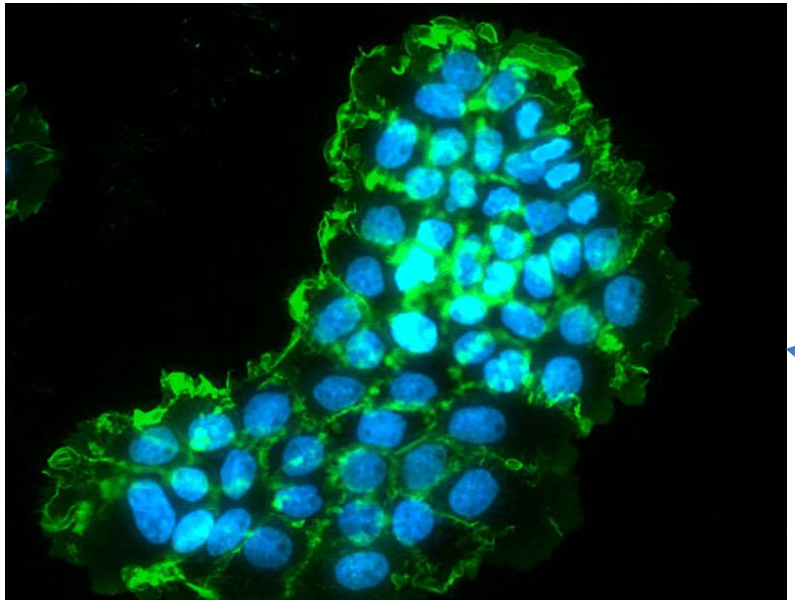
Robustness Repeatability

Quantitation Accuracy (how much?)

# Microscopy: Immunohistochemistry, immunofluorescence

IHC: Targeted ntibody reacts with somatostatin 2a receptors over-expressed in insulinoma (300x, H&E counterstain, reaction is RED)





A431 epithelial cc. tumor cells, nuclei stained with Hoechst-blue, PHOSPHORILATED EGF receptors in cell membrane are GREEN using an antibody coupled to 488 nm emission DyLight fluorescent stain

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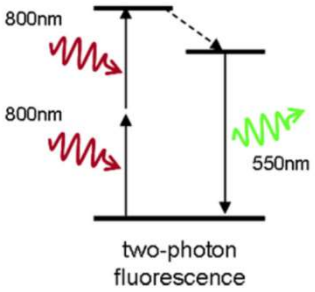
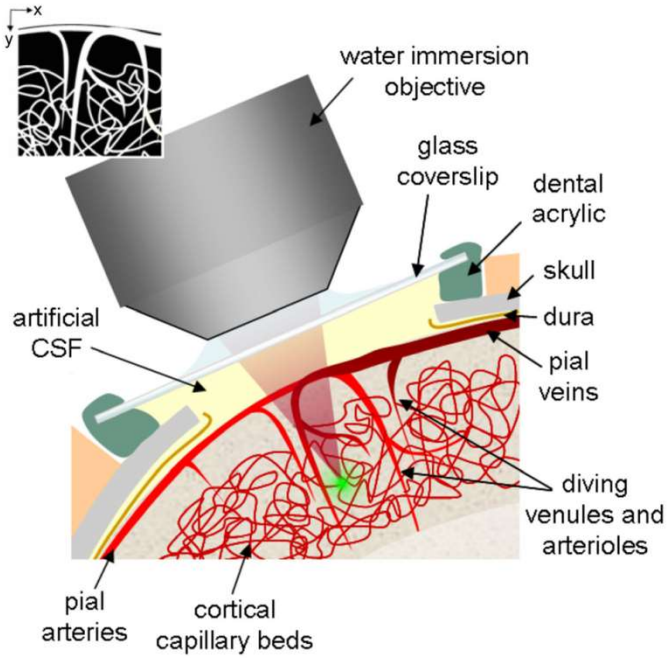
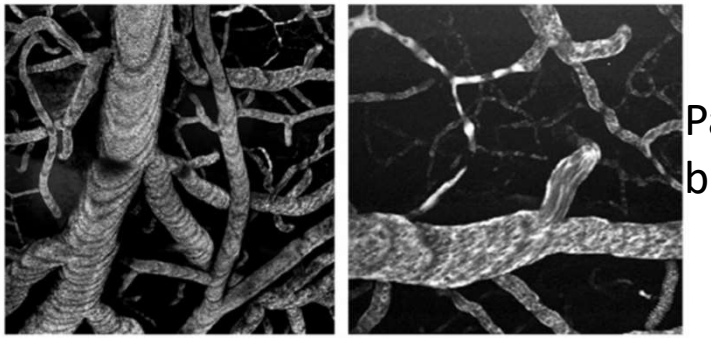
# High definition methods in live animals: in vivo dual photon microscopy

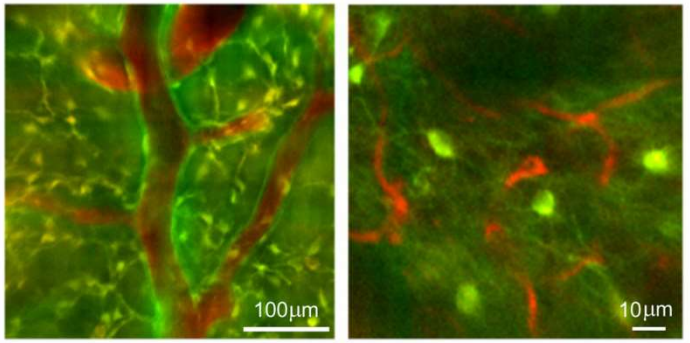
better S/N ratio
increased resolution



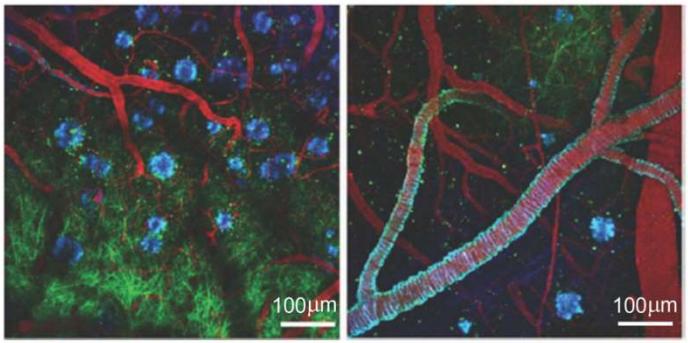



Pattern of RBC flow in brain vessels




Vessels and neurons (dextran Texas Red and GFP-TG mouse)




Amiloid plaques (Blue), Vessels (red) neurons(green)

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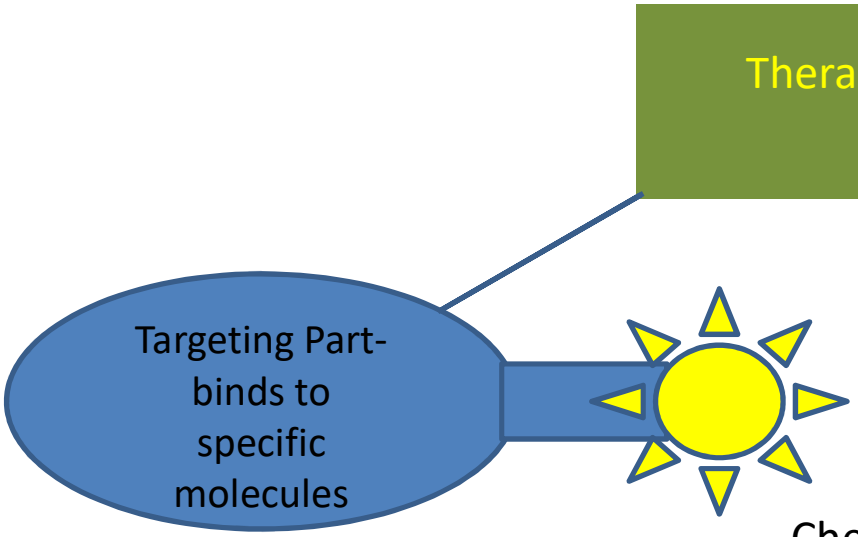
# General Structure of a Molecular Probe



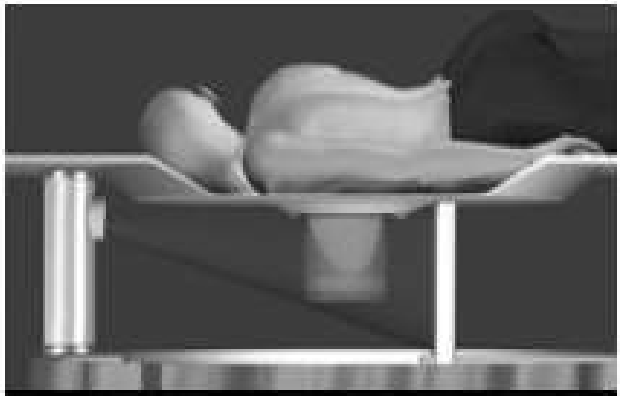

Signalling/Contrasting Element

Small Molecules
Peptides
Proteins/Subunits
Antibodies/Subunits

<u>Chemical Nature:</u>
PET/SPECT: isotopes (radioactivity)
Optical/Acoustic: Fluorescent Dyes
MRI: Gd, Fe
CT: iodine, Barium sulphate
<u>Nano/microparticles:</u>
Optical: quantum dot, carbon nanotubes,
Au particles
MRI: iron and Mn-oxide particles
CT: golden particles

| Modalitás | Előnyei | Hátrányai | Fontos kontrasztanyag/jel | Klinikai alkalmazás példái |
|---|---|---|---|---|
| CT | Any image depths<br>Good time resolution<br>Perces kép-idők<br>Közepesen drága<br>Anatómiai módszer | Sugárterhelés<br>Rossz lágyszöveti kontraszt<br>Jelenleg csak anatómiai és funkcionális képalk. | Ba, I, Kr, Xe | Tumor perfúzió, |
| PET | Bármilyen mélységű kép<br>Egésztest-képalkotás<br>Kvantitatív mérések<br>Kombinálható CT/MRI-vel | Sugárterhelés<br>Drága<br>Milliméteres felbontás<br>Hosszabb képidő (perc-óra) | C-11, F-18, Ga-68, Cu-64, Zr-89 | FDG-PET tumor staging, különböző betegségek diagnosztikája |
| SPECT | Bármilyen mélységű kép<br>Egésztest-képalkotás<br>Kvantitatív mérések<br>Multiplex<br>Teragnosztika<br>Kombinálható CT-vel | Sugárterhelés<br>Szubmilliméteres-milliméteres felbontás<br>Hosszabb képidők | Tc-99m, I-123, In-111, Lu-177 | Molekuláris diagnosztika<br>Radioterápia (NHL, NET, pm. cc.) |
| MRI | Bármilyen mélységű kép<br>Egésztest-képalkotás<br>Nincs ionizáló sugárzás<br>Kitűnő lágyszöveti kontraszt | Drága<br>Hosszú képidők<br>Korlátozott érzékenység | $Gd^{3+}$, vas-oxid részecskék (SPIO, USPIO) | Prosztata daganat nycs. met.<br>Fokális májléziók<br>Szív perfúzió |
| MRS | Nincs ionizáló sugárzása<br>Egésztest-képalkotás | Drága<br>Hosszú képidők<br>Kis érzékenység | Kolin, laktát, kreatin, lipidek, N-acetil-aszpartát | Agytumorok anyagcseréje<br>Alzheimer-kór követése |
| UH | Nincs ionizáló sugárzás<br>Rövid/valósidejű képalkotás<br>Nagy térbeli felbontás<br>Olcsóság<br>Nagy érzékenység | Egésztest-képalkotás nincs<br>Kontrasztanyagok csak az érrendszerre<br>Operátor-függő | Mikro-buborékok | Fokális májléziók, echokardiográfia,<br>Tumor perfúzió |
| Optikai módszerek | Nincs ionizáló sugárzás<br>Rövid/valósidejű képalkotás<br>Nagy térbeli felbontás<br>Olcsóság<br>Nagy érzékenység, kvantitatív<br>Multiplex | Korlátozott áthatolóképesség (1 cm)<br>Nincs egésztest-képalkotás | Fluoreszcens molekulák és festékek, fény-elnyelő nanorészecskék | OCT-érelmeszesedés, retinopathiák, kolonoszkópia |

| CT | 3D Attenuation Map of X-Rays |
|---|---|
| Advantages | Any Imaging Depth<br>Good Resolution<br>Simple<br>Medium-Priced<br>Sub-minute scan times |
| Disadvantages | Radiation Dose<br>Soft Tissue C.<br>Just anatomical and functional images<br>Any imaging depth<br>Good Resolution<br>WB Imaging<br>Minute Scanning Time<br>Semi-Expensive<br>Anatomy |
| Contrast Materials | Ba, I, Kr, Xe, Au |
| Clinical Use | Tumor perfusion, Ca-score, Ventillation |
| Voxel Sizes, Cells Per Voxel | 1x1x1 mm<br>1 million |

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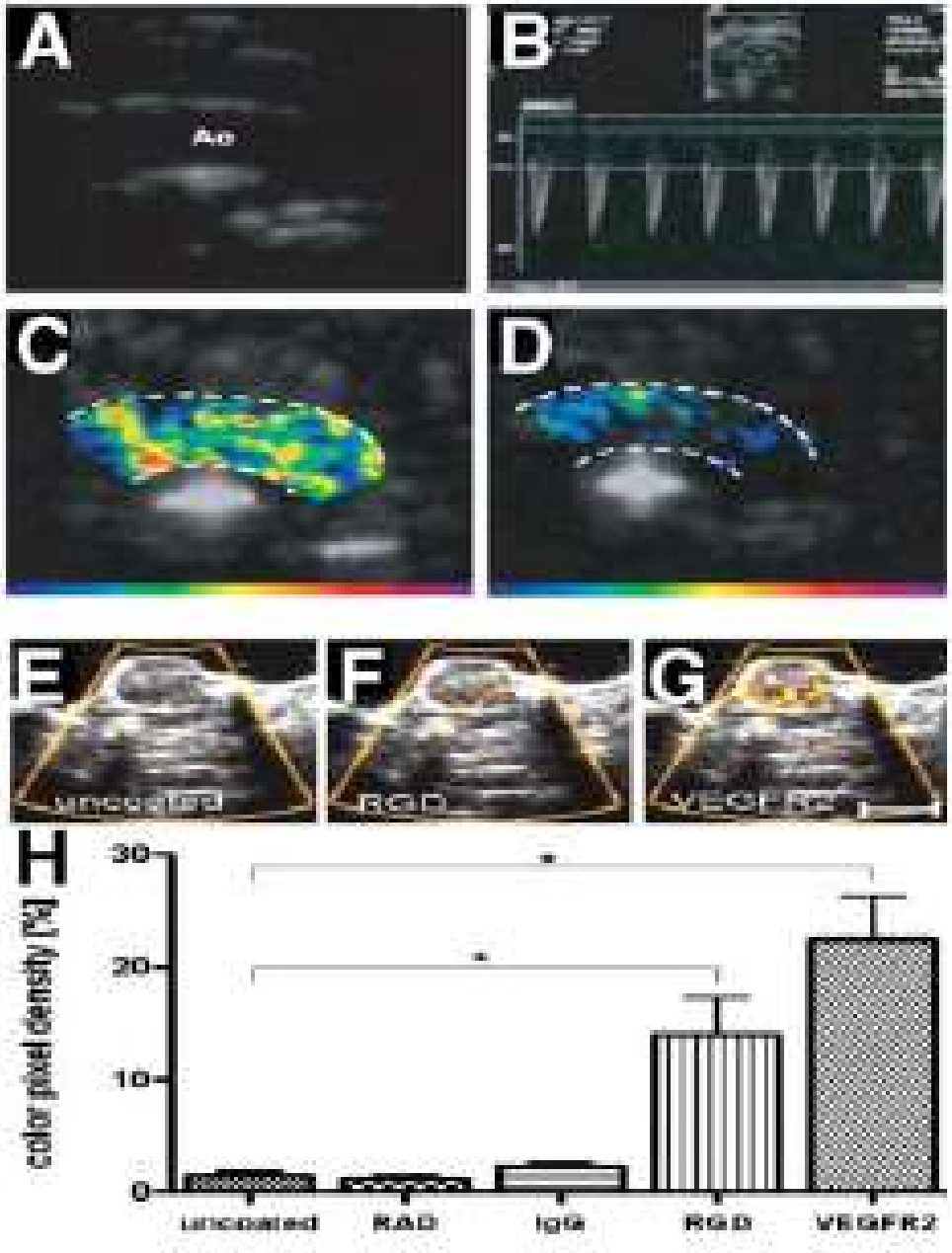
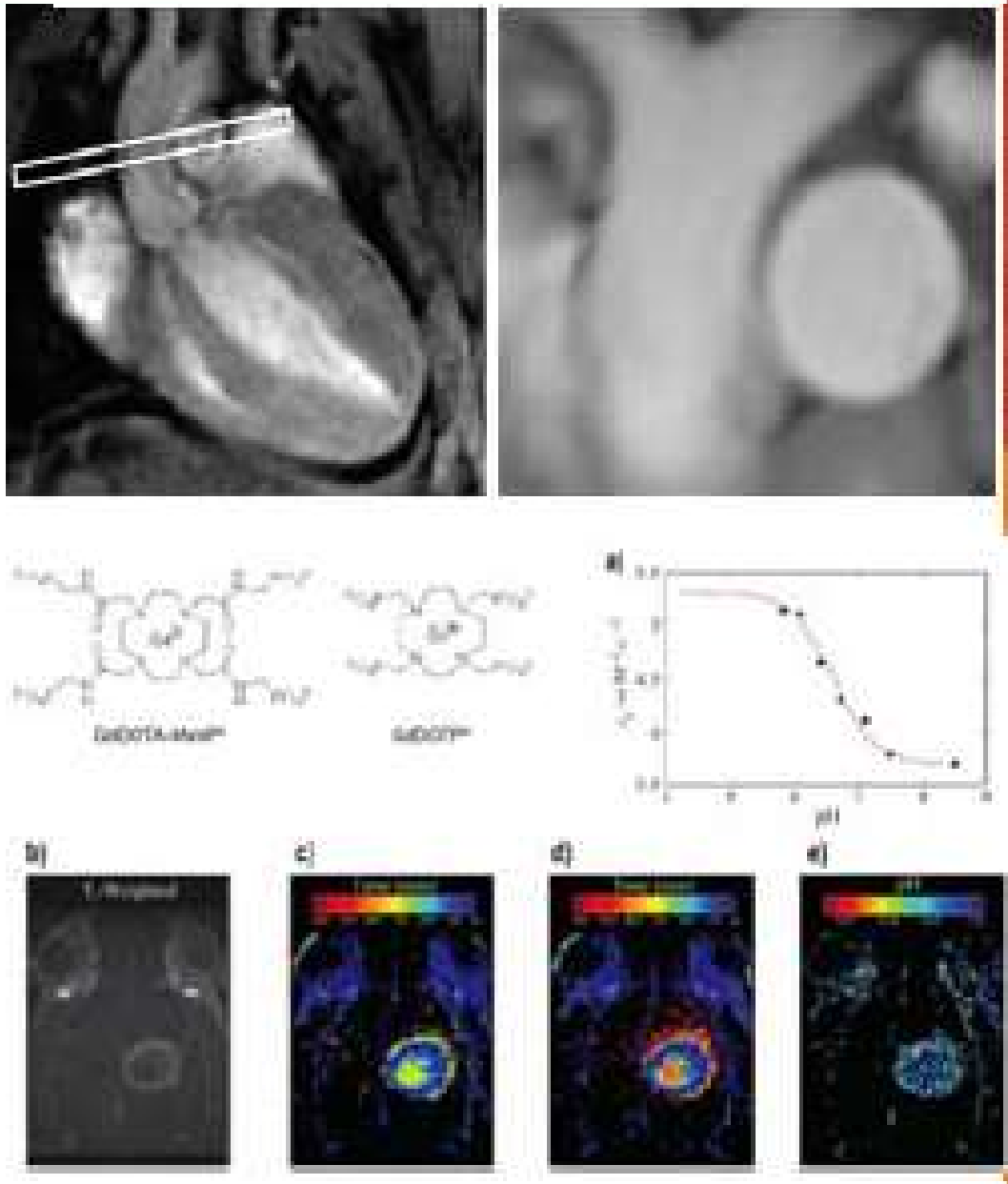
| UltraSound | 3D differences in echo and spreading of sound waves |
|---|---|
| Advantages | No ionizing radiation<br>Fast/RT imaging<br>High Sensitivity<br>High Resolution<br>Cheap |
| Disadvantages | No WB img.<br>Only vascular contrast materials<br>Operator dependency |
| Contrast Materials | Micro-Bubbles |
| Clinical Use | Focal liver lesions, Echocardiography, Blood perfusion…Rare, Prostate Cancer VEFG Expression (Phase III BR55) |
| Voxel Sizes, Cells Per Voxel | 1x1x1 mm<br>1 million |



| MRI (IRM…) | 3D / 2D Image of Proton Spins |
|---|---|
| Advantages | Any Imaging Depth and Plane<br>WB Imaging<br>No ionizing radiation<br>High Soft Tissue Contrast |
| Disadvantages | Expensive<br>Low Sensitivity<br>Long imaging time |
| Contrast Materials | Gd3+, iron-oxide particles (SPIO, USPIO) |
| Clinical Use | Liver, Brain Lesions, Cardio-MRI |
| Voxel Sizes, Cells Per Voxel | 1x1x1 mm<br>$10^{13}$ |



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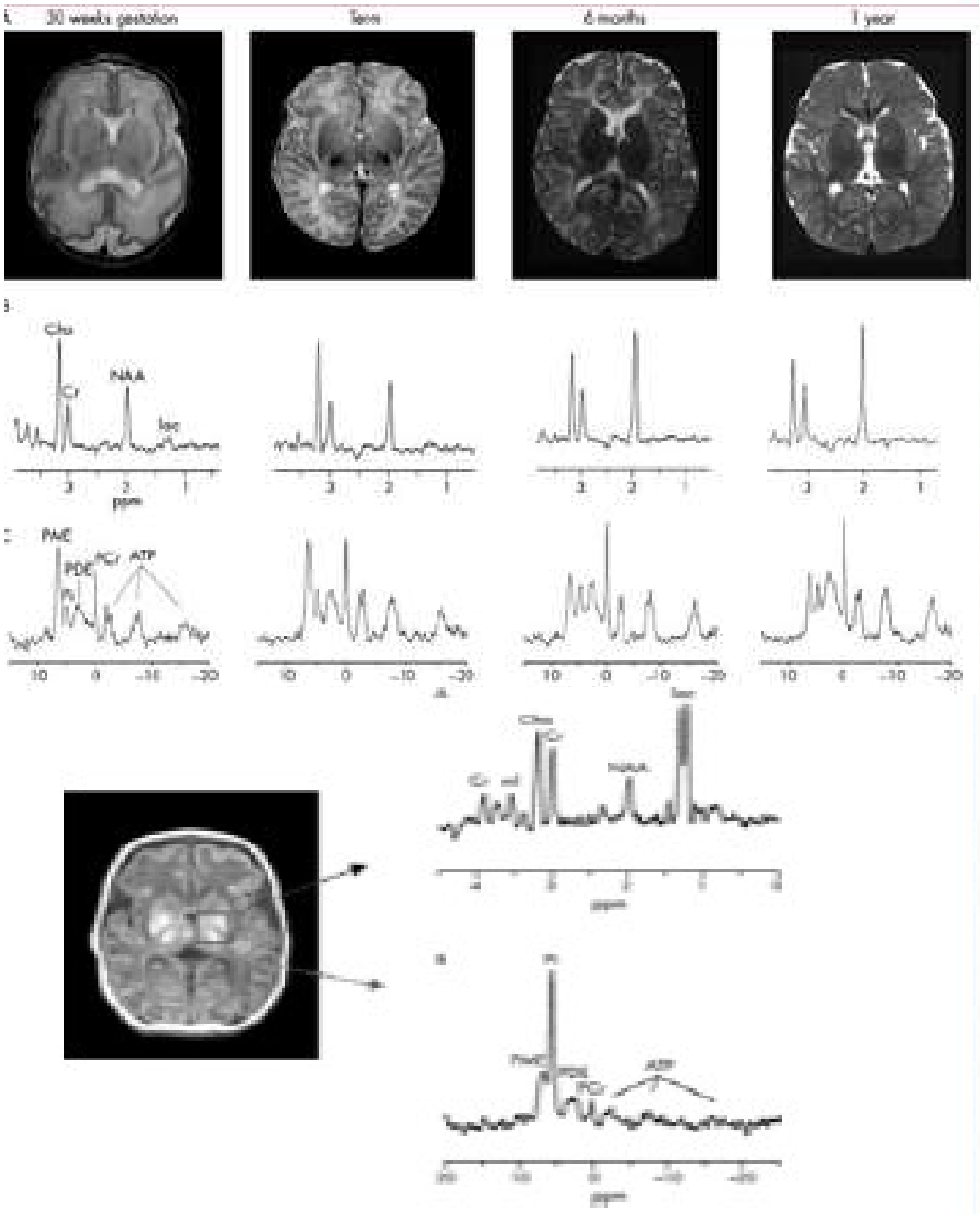
| MRS | Recorded Electromagnetic Resonance Spectrum |
|---|---|
| Advantages | No ionizing radiation<br>WB Imaging |
| Disadvantages | Expensive<br>Very low sensitivity<br>External Calibration / Special Knowledge |
| Contrast Materials | Cholin, Lactate, Creatin, Lipids, N-Ac-Aspartate |
| Clinical Use | Brain Tumor Stratification, Stroke |
| Voxel Sizes, Cells Per Voxel | N.A. |

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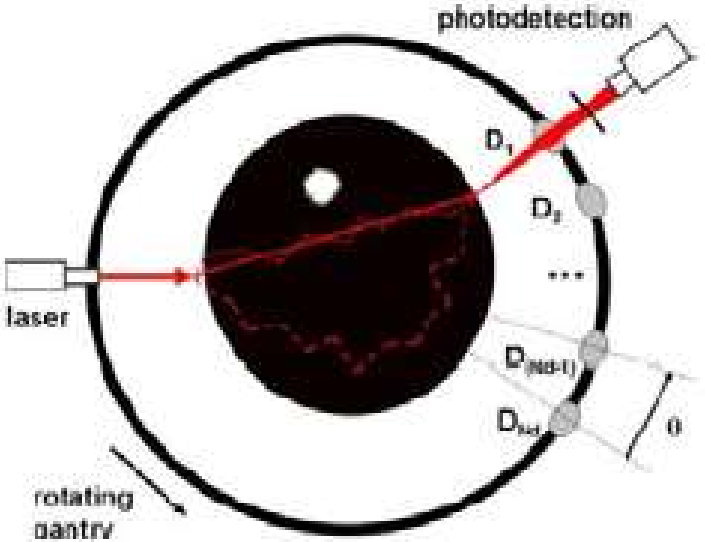
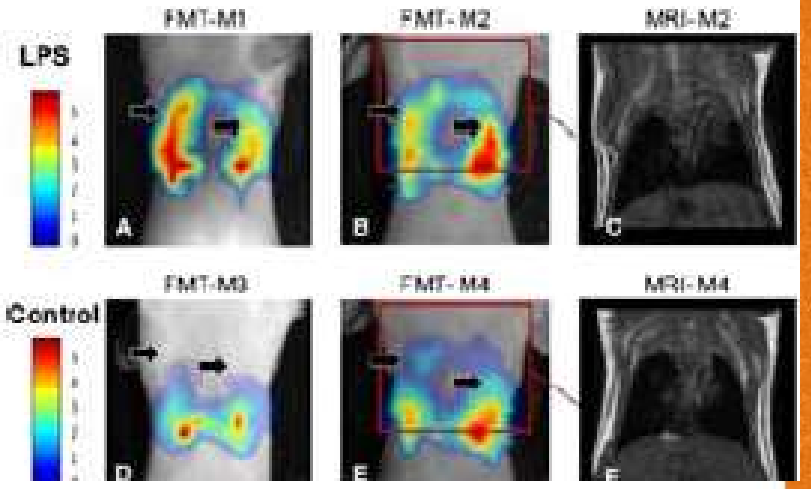
| Optical Methods | Visible/NIR Light Transmission/Reflectance/Emission/ Scatter in 2D , 3D |
|---|---|
| Advantages | No ionizing radiation<br>Short/RT Imaging<br>High Spatial Resolution<br>Very Sensitive, Semi-Quantitative<br>Multiplex |
| Disadvantages | Limited Transparency<br>No WB imaging |
| Contrast Materials | Fluorescent molecules, Light-emitting reactions, Dyes, QD-s, NP-s |
| Clinical Use | Experimental, Sentinel Ln., Image-Guided Surgery, Retinopathies (OCT), Mammary screnning (LumaGem) |
| Voxel Sizes, Cells Per Voxel | 2D: 0.01 mm2<br>3D: 0.8x0.8x0.8 cm<br>$10^3$ (2D), $10^{13}$ (3D): cca.$10^{4-5}$ per cell |

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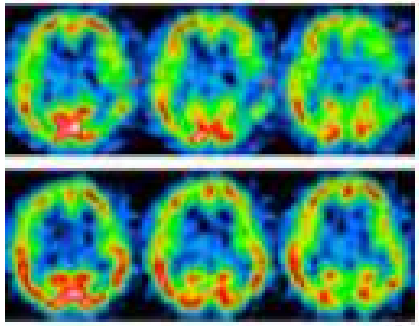
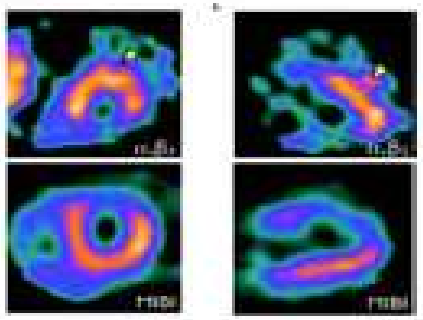
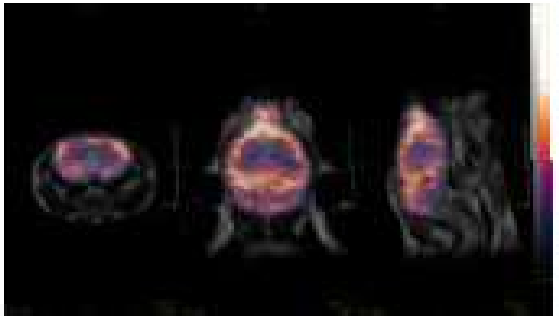
| SPECT | 3D Gamma-ray Source Array in Tissue |
|---|---|
| Advantages | Any imaging depth<br>WB Imaging<br>Quantitative<br>Good resolution<br>Multiplexing<br>Theragnostics<br>Combination w/CT |
| Disadvantages | Radiation Dose<br>Sub-mm Resol.<br>Long imaging times |
| Contrast Materials | Tc-99m, I-123, In-111, Lu-177, Ho-166, Tl-201 |
| Clinical Use | Nuclear Cardiology, Brain Perfusion, Oncology (AB, Peptides), Receptor T. |
| Voxel Sizes, Cells Per Voxel | Clinical: 0.8 x 0.8 x0.8 mm<br>Small A: 0.3 x 0.3 x 0.3 mm<br>2 pM / voxel<br>1/10 atom per cell |



I-123 iomazenil agyi SPECT



Tc-99m-MIBI szív perfúzió + angioneogenezis SPECT I-123-integrin liganddal



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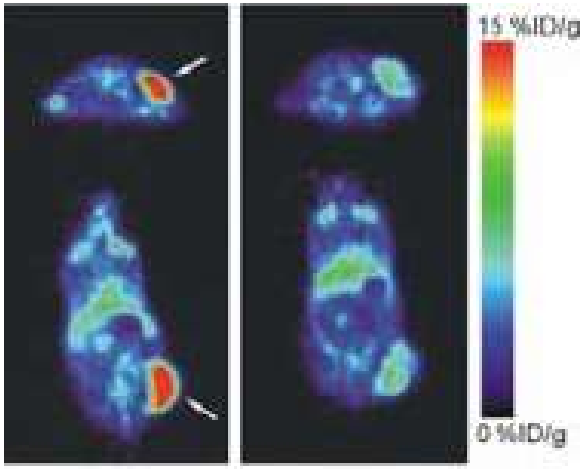
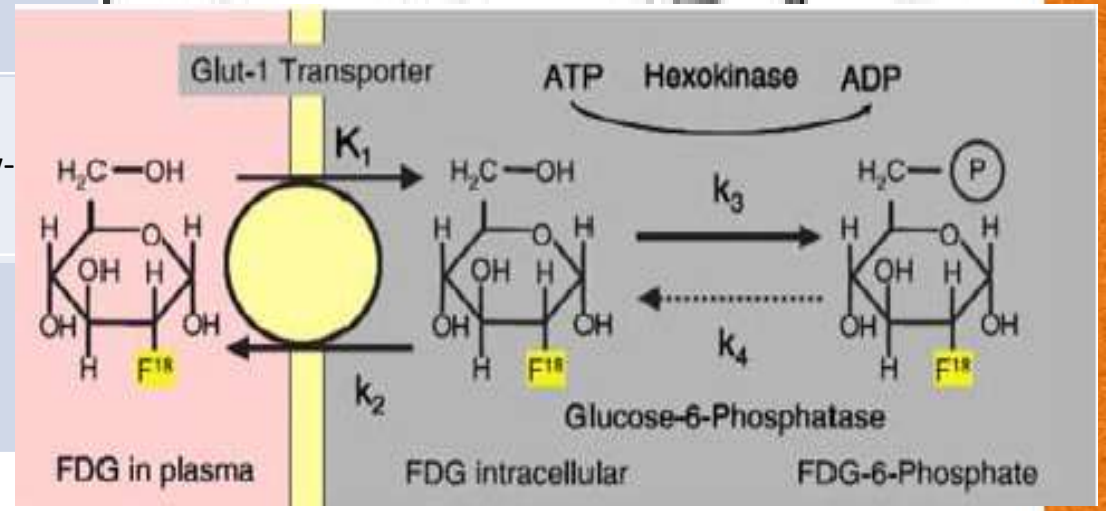
| PET | 3D 511 keV Gamma-ray Source Array in Tissue |
|---|---|
| Advantages | Any imaging depth<br>WB Imaging<br>Quantitative<br>Anatomical Co-registration CT, MRI |
| Disadvantages | Radiation Dose<br>PRICE<br>Resolution<br>Longer imaging times |
| Contrast Materials | C-11, F-18, Ga-68, Cu-64, Zr-89 |
| Clinical Use | FDG Patient Staging+Follow- |
| Voxel Sizes, Cells Per Voxel | 5 x 5x 5 mm<br>0.6 x 0.6 x 0.6 mm<br>0.02 pM/voxel<br>1/100 atom per cell |

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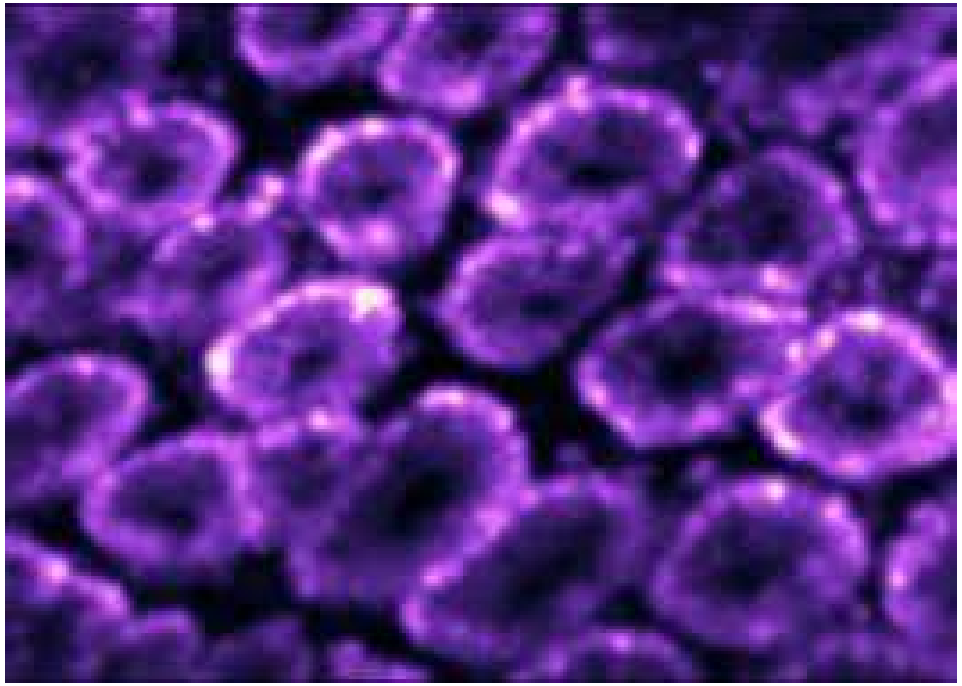
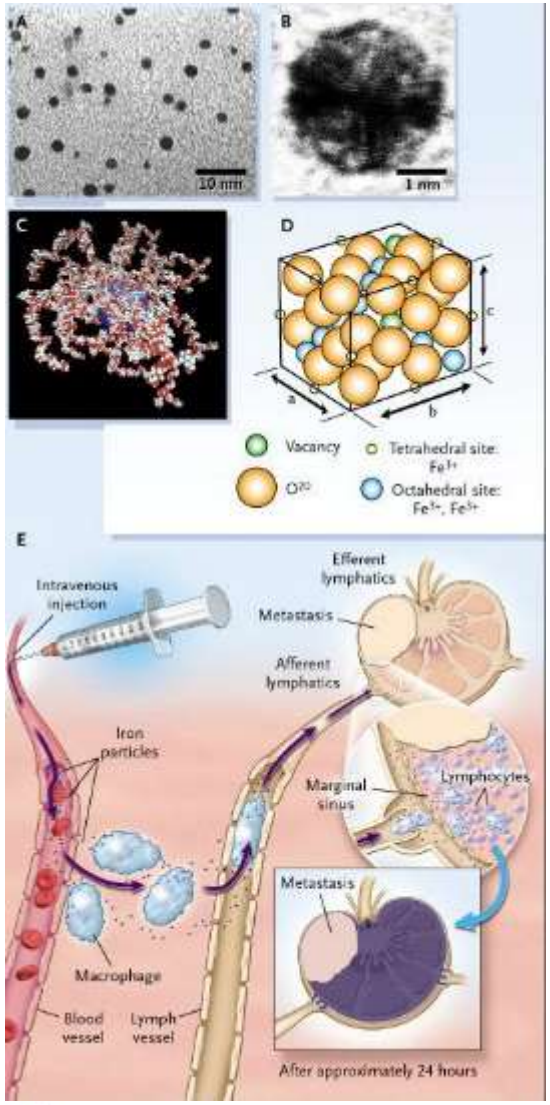
# Screening-Confocal Endomicroscopy

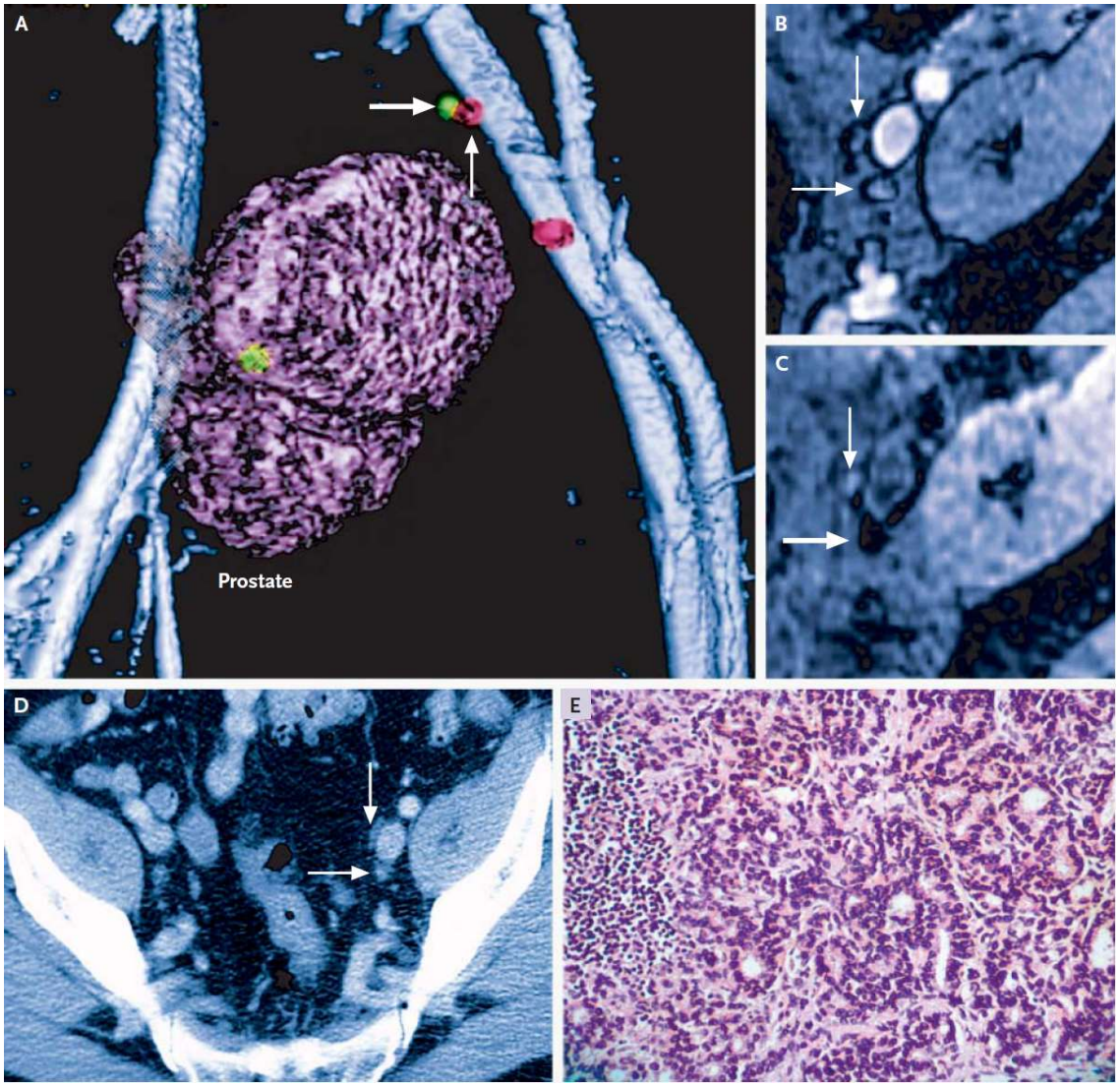

Oesophagus, stomach, bile ducts, ileal/colonic mucosa examined on cell level using fiber optic confocal microscope at autofluorescence emission/excitation wl-s

In vivo real time image of a patient's colonic crypts



# Diagnostics-USPIO MRI-A SAD STORY




Harishingani M et al N.Engl. J:Med. 2003

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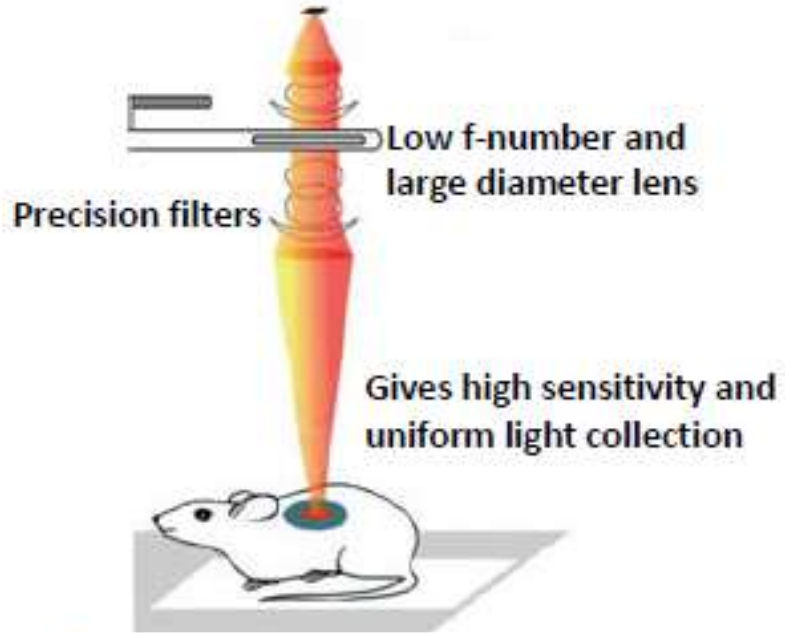
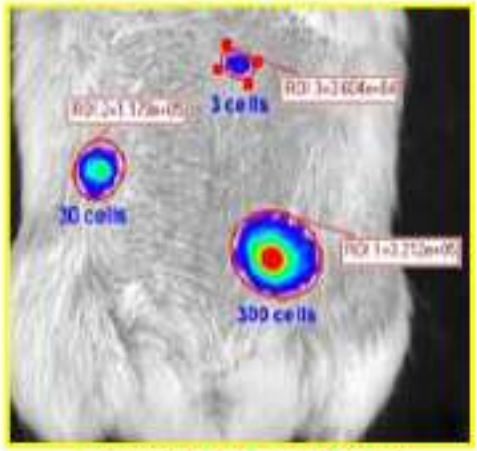
BIOLUMINESCENE-The power of nature helping to collect IF-s
Firefly, Jellyfish – TG animals

**Cooled (-90C) camera with large CCD chip area for high sensitivity**




**Resolves multiple bioluminescent reporters**
**Even detects single cells *in vivo***



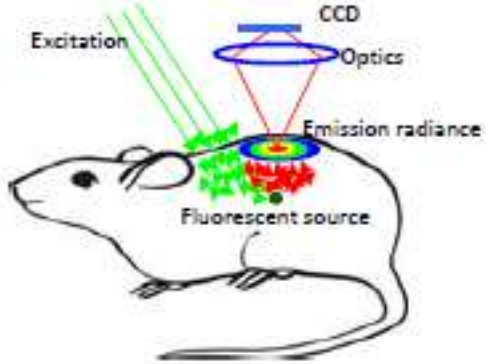

Rabinovich *et al.* (2008)
PNAS 105(38): 14342-6

In vivo imaging of s.c. implanted T cells transduced with optimized firefly luciferase (left) and a 'single' 4T1 breast cancer cell (right)

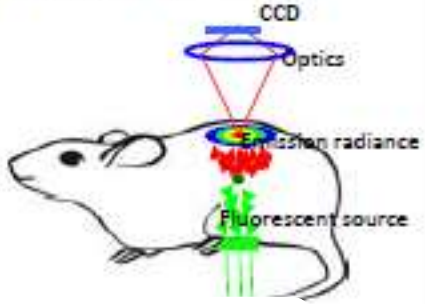
# FLUORESCENCE IMAGING

**Cooled (-90C) camera with large CCD chip area for high sensitivity and a choice of imaging modes for maximal flexibility, *e.g.*, transmission for deep tissues.**

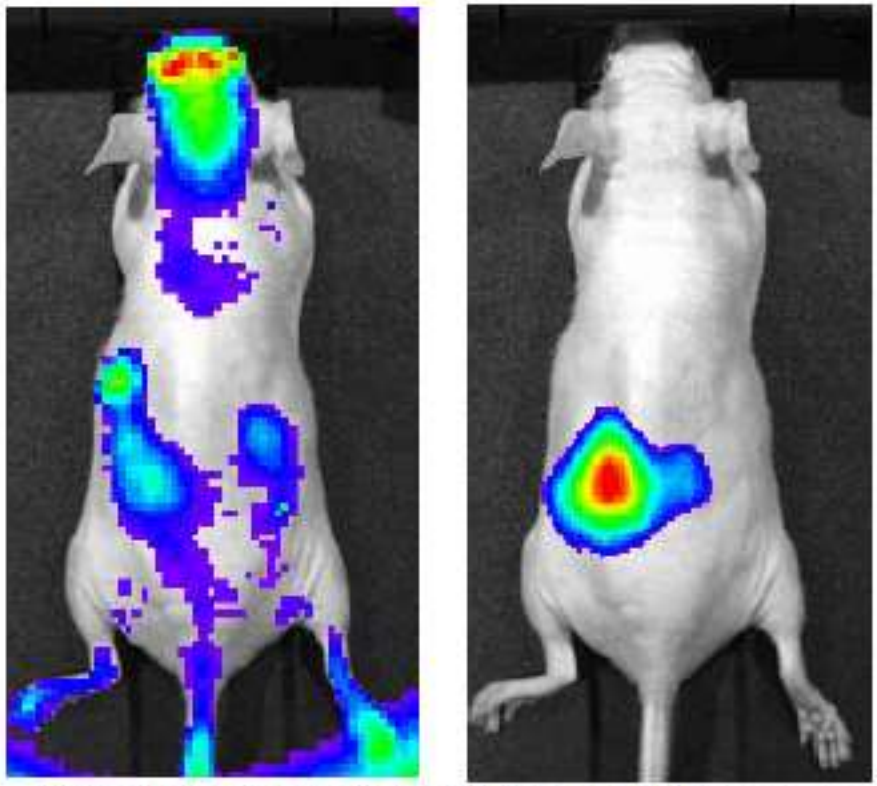
## Reflectance




## Transmission

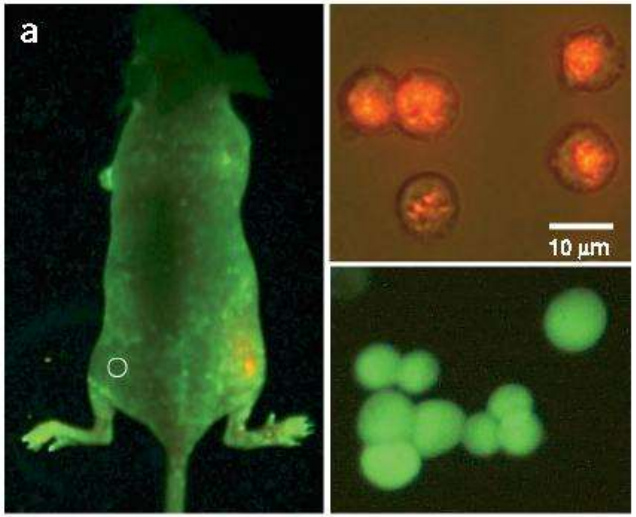


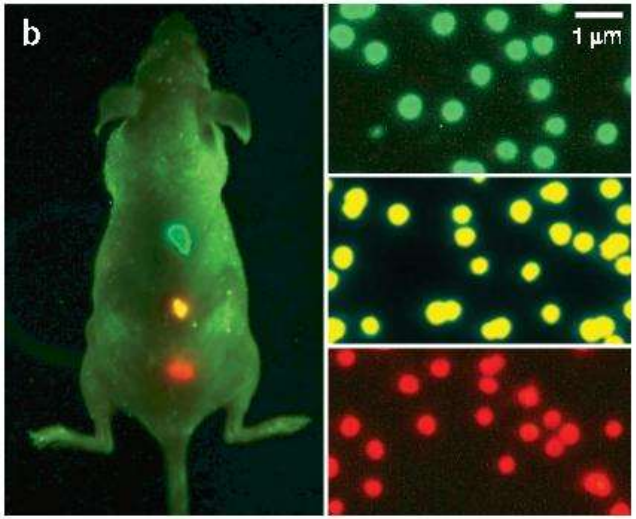

**Epi-Illumination** **Transillumination**

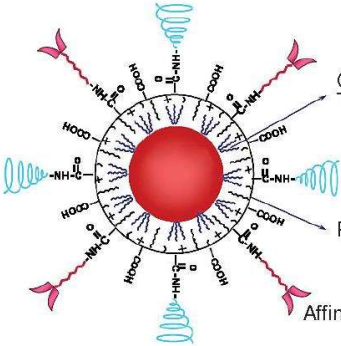


Pillow implanted medial to left kidney, 1x1015 molecules

# Nanobiotechnológiai és In Vivo Képalkotó Központ



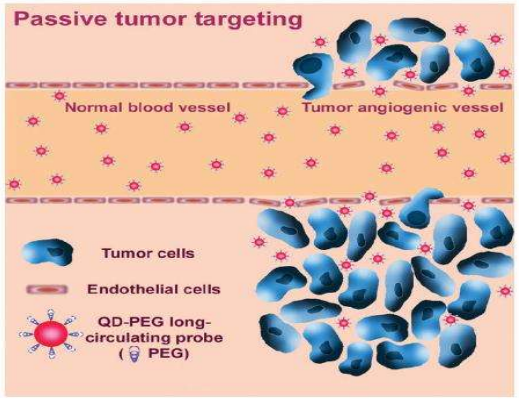



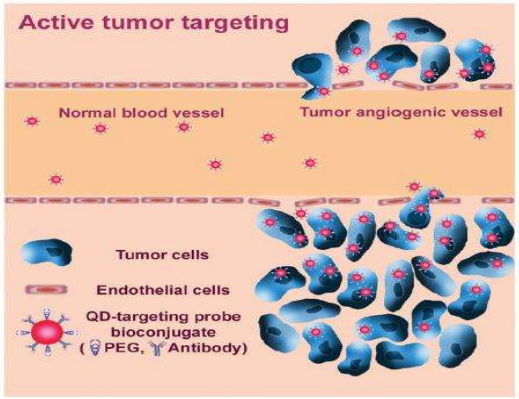




QD capping ligand: TOPO O=P

PEG: poly (ethylene glycol) $-(CH_2-CH_2-O)_n-$ MW = 5,000

Affinity ligands: antibody, peptide, small-molecule drug, inhibitors

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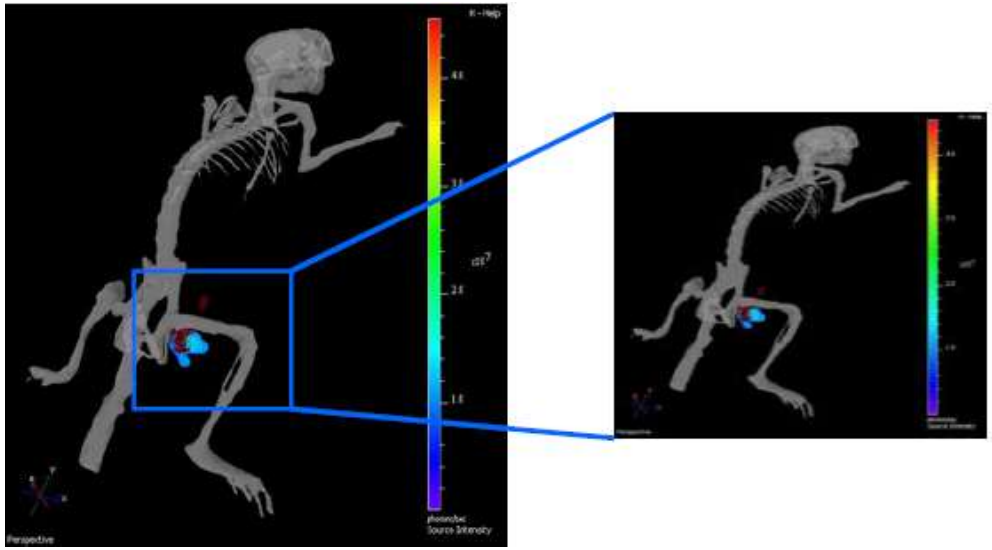
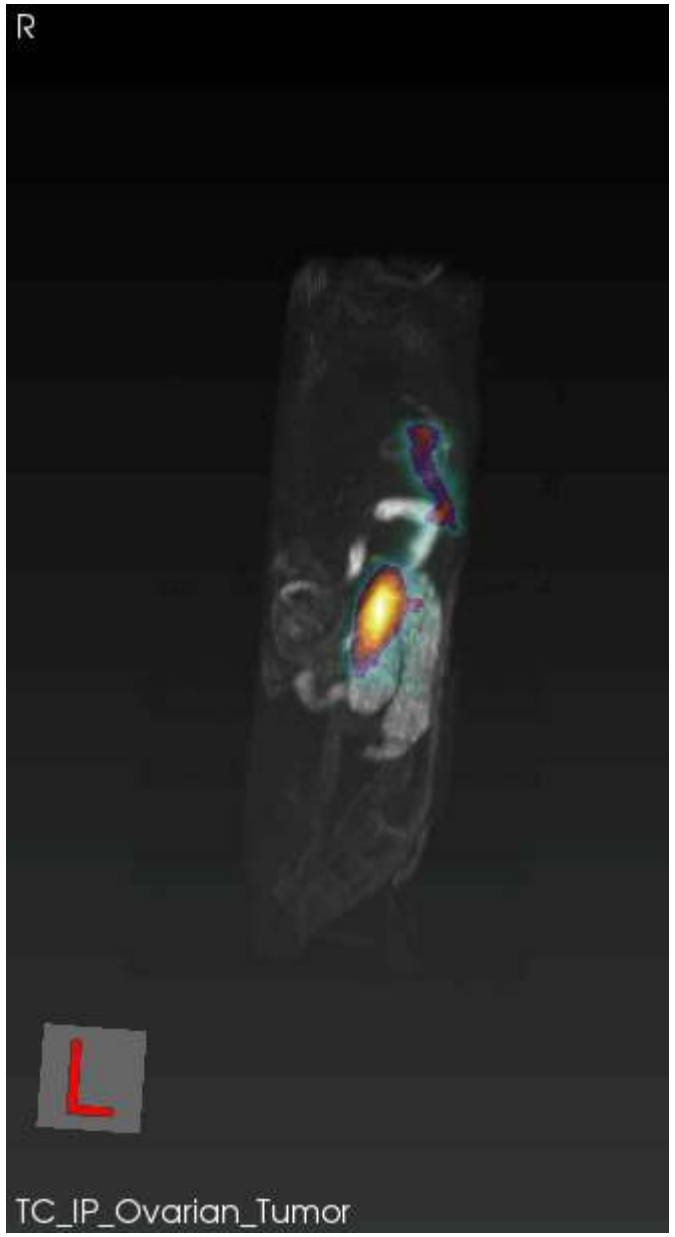
# Nanobiotechnology and In Vivo Imaging Center

# 3D Optical Tomography



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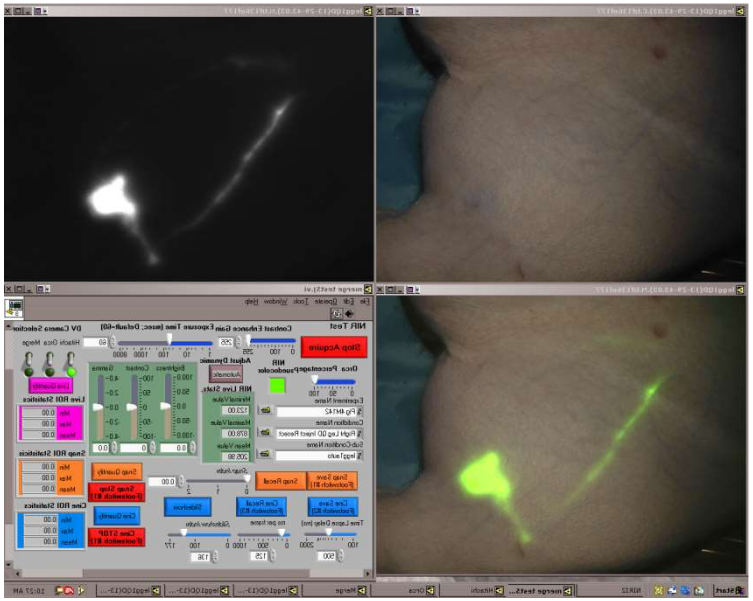
# Image guided surgery



John Frangioni FLARE Harvard

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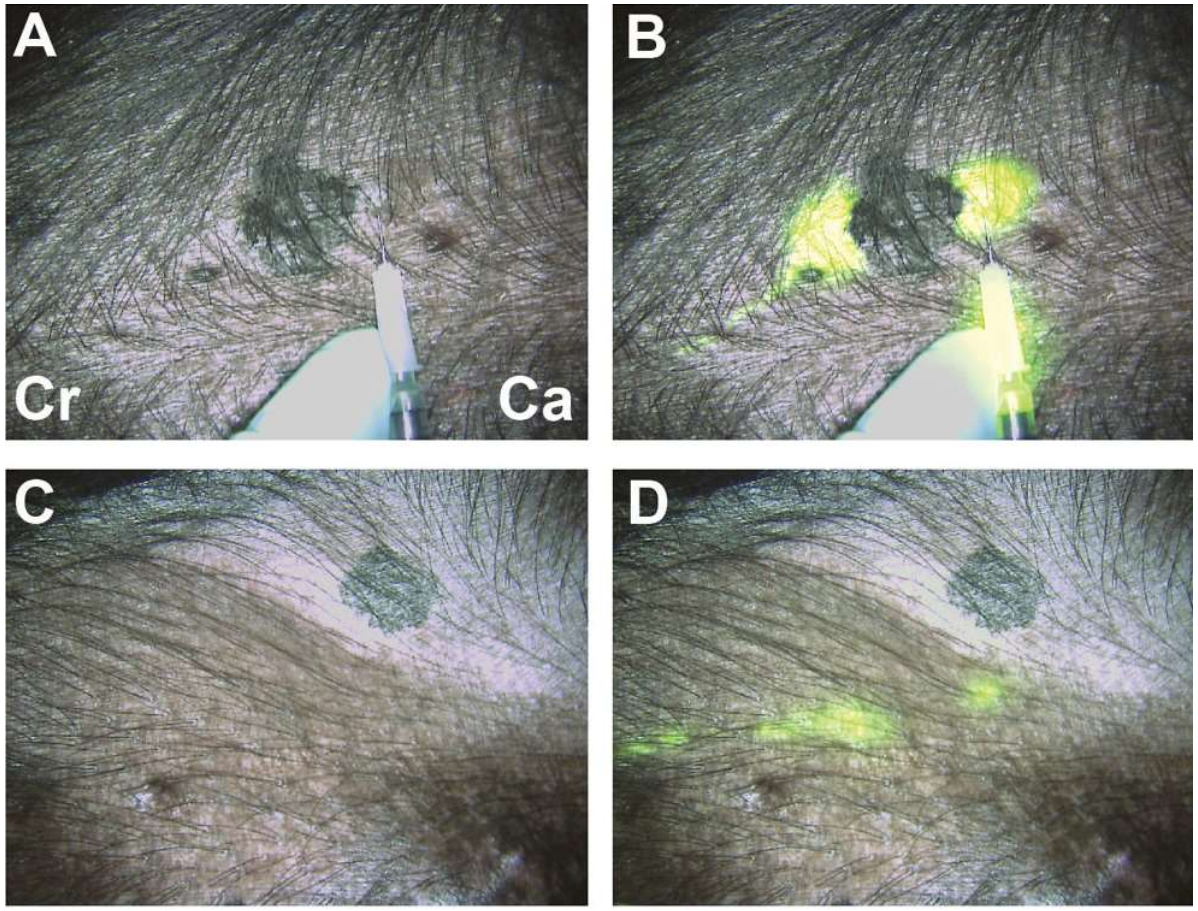
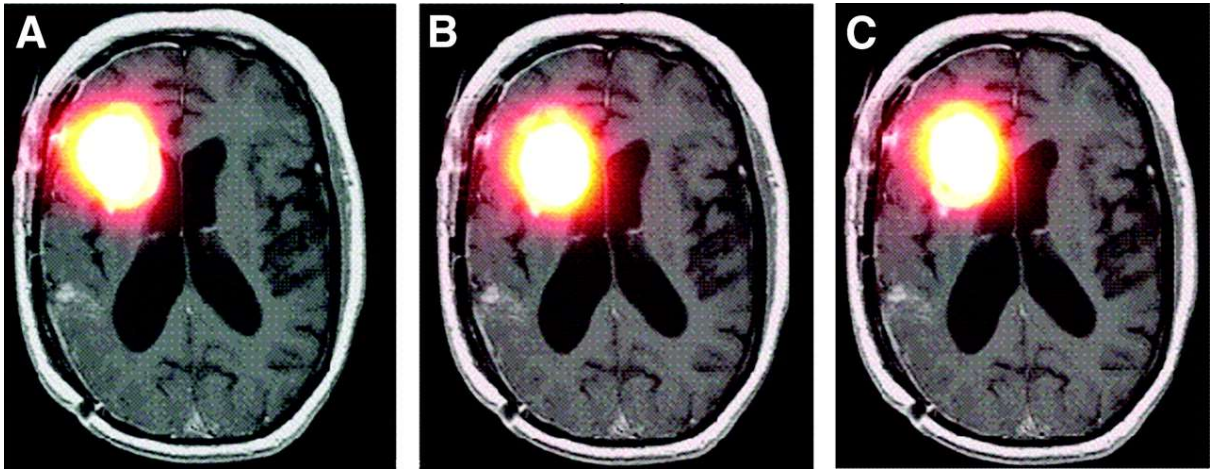
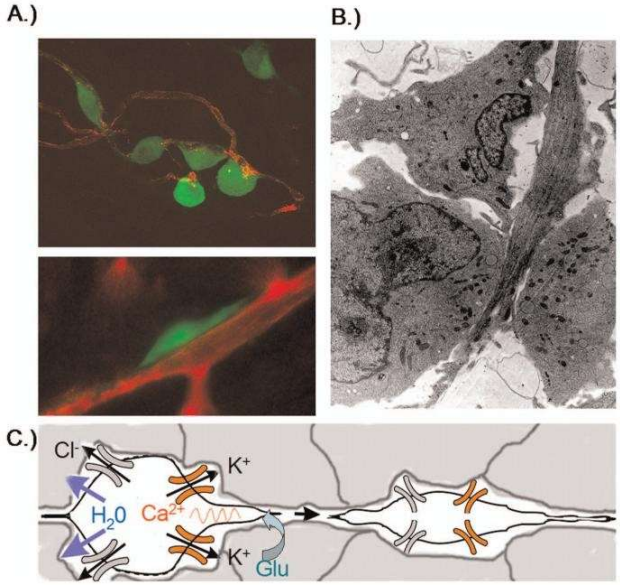
# Planar Fluorescence In the Clinic

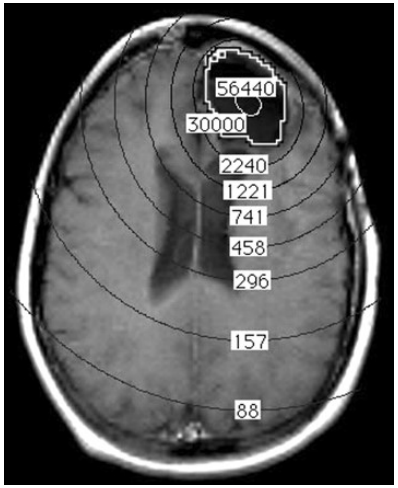



Melanoma, ICG-Albumine particle

# Personalized Targeted Therapy: SPECT/MRI/RNT

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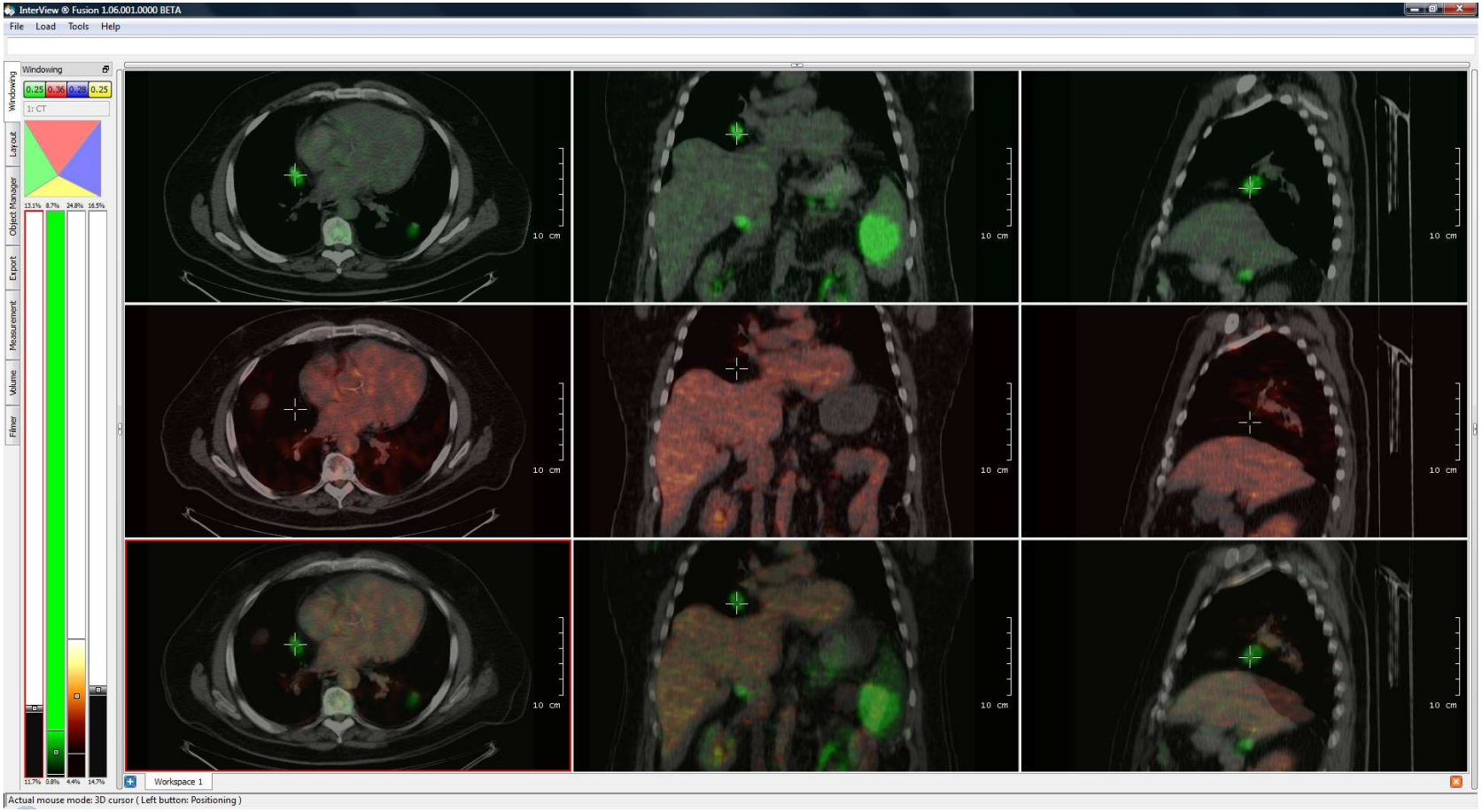
# Monitoring with FDG-PET

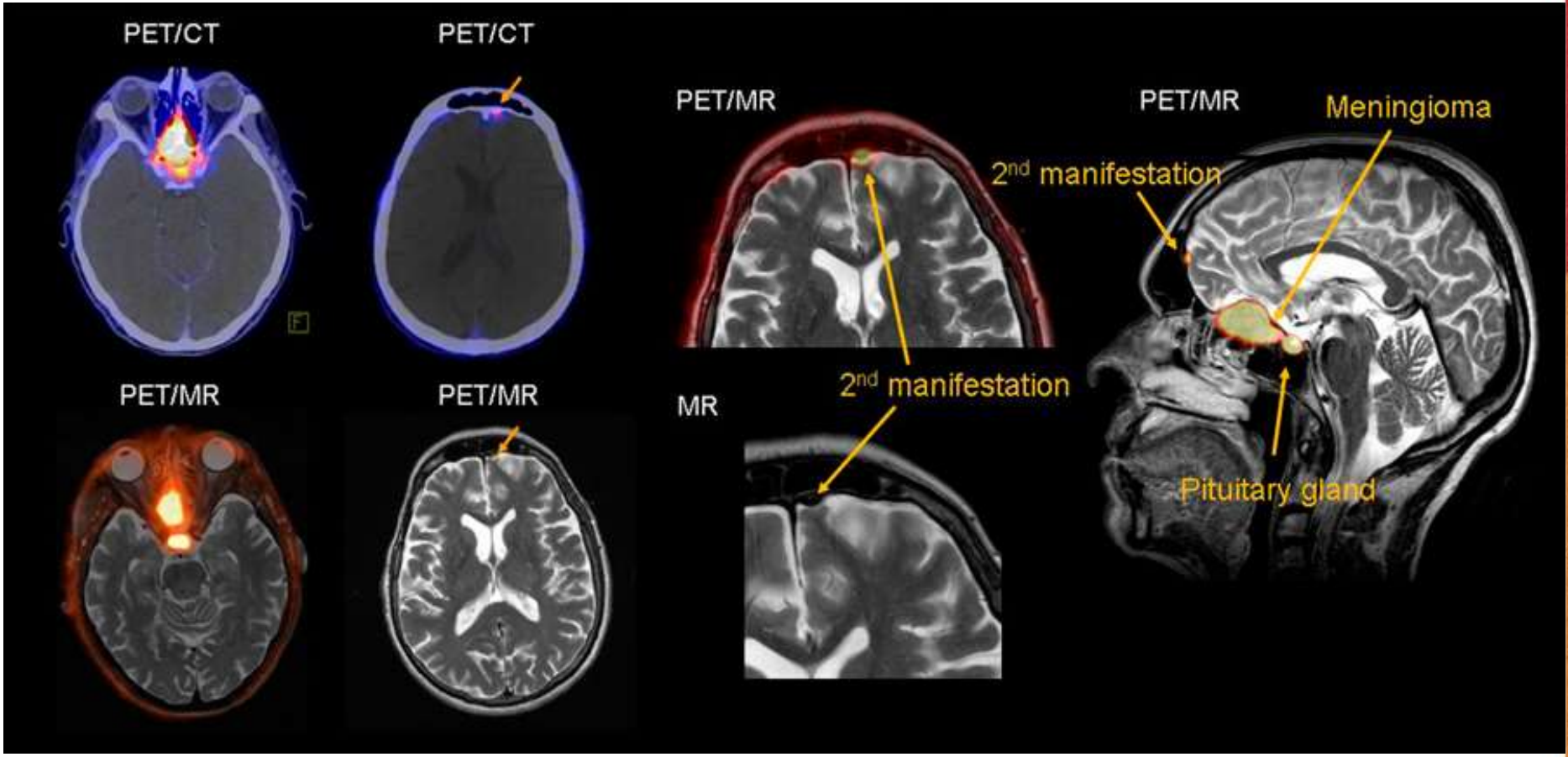



1st PET-CT

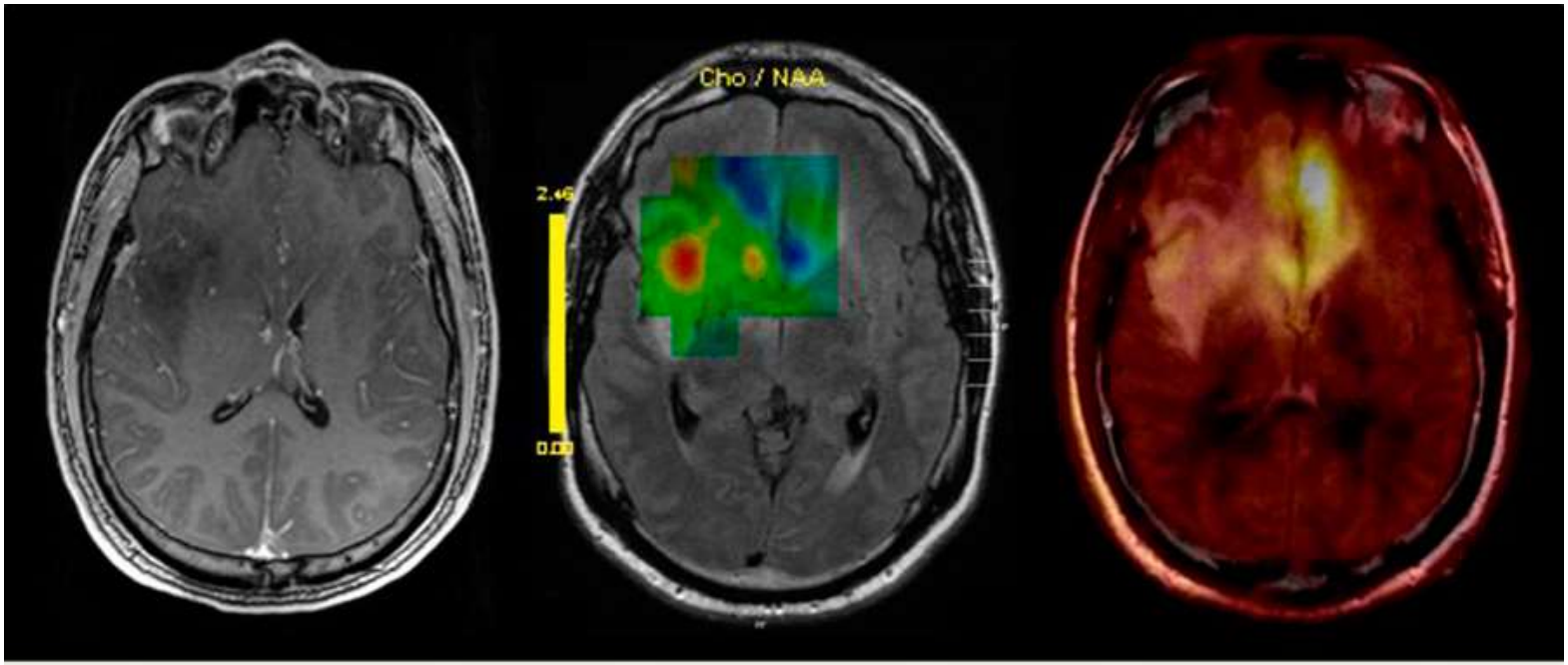
2nd PET-CT

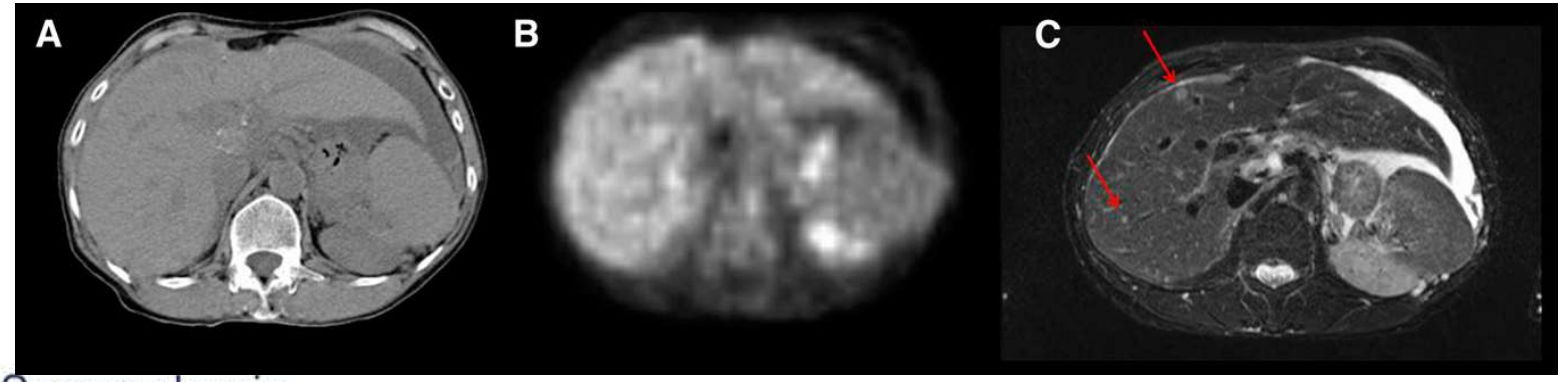
Quadruple fusion

# PET/MRI

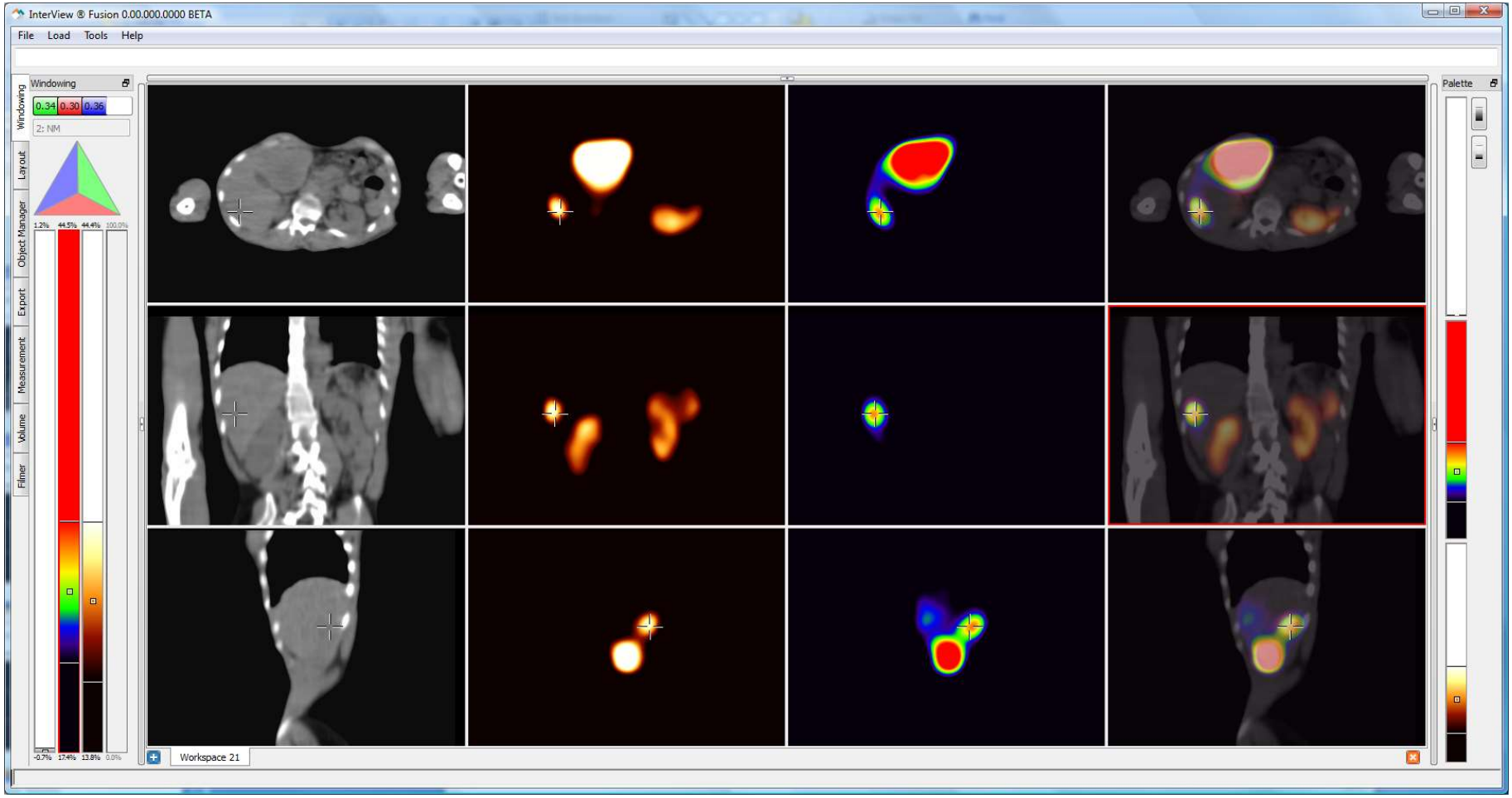

PET/CT
PET/CT
PET/MR
PET/MR
Meningioma
2nd manifestation
2nd manifestation
MR
PET/MR
PET/MR
Pituitary gland

# PET/MRI

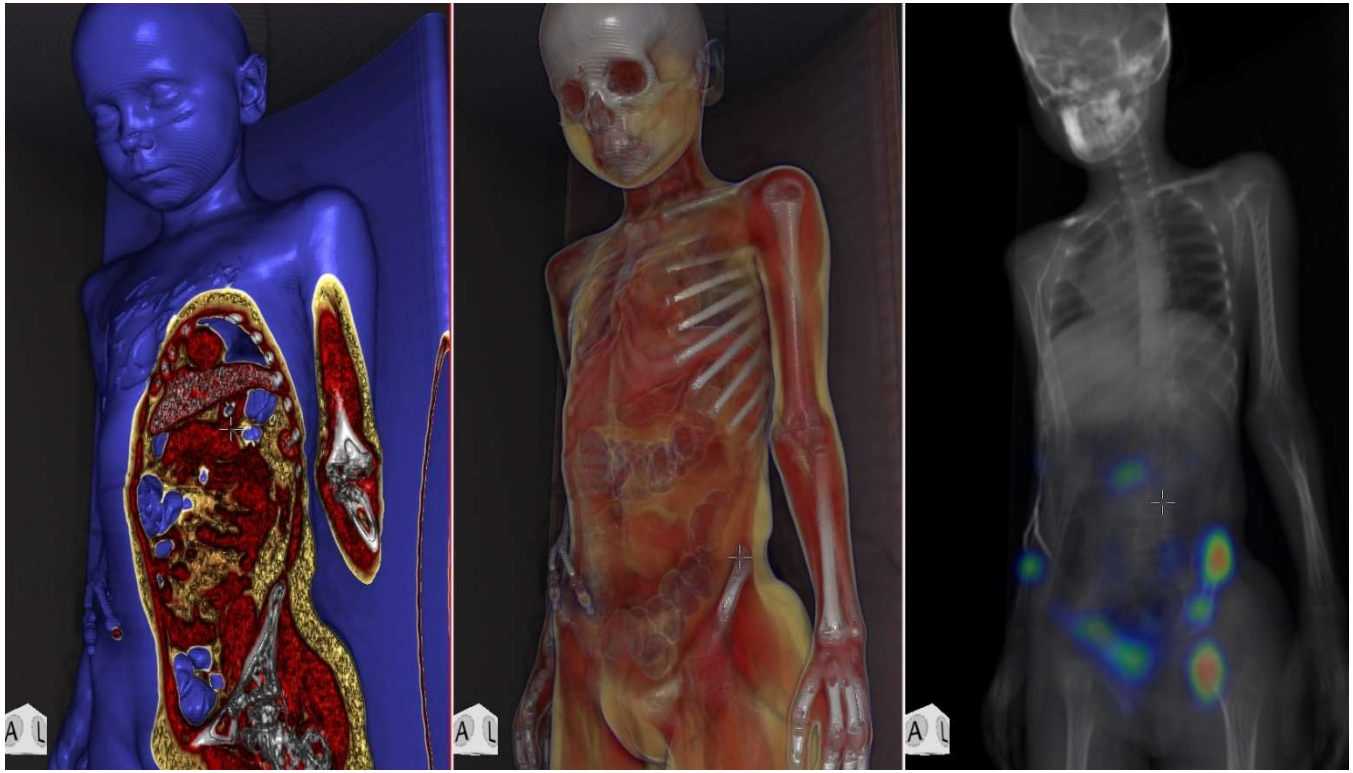


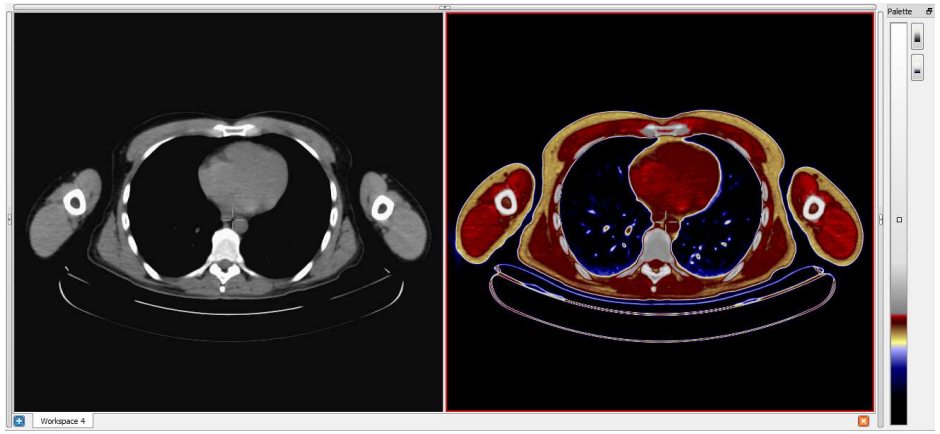



# SPECT for neuro-endocrine tumor treatment/assessment



# SPECT-CT look-up table (color scale)



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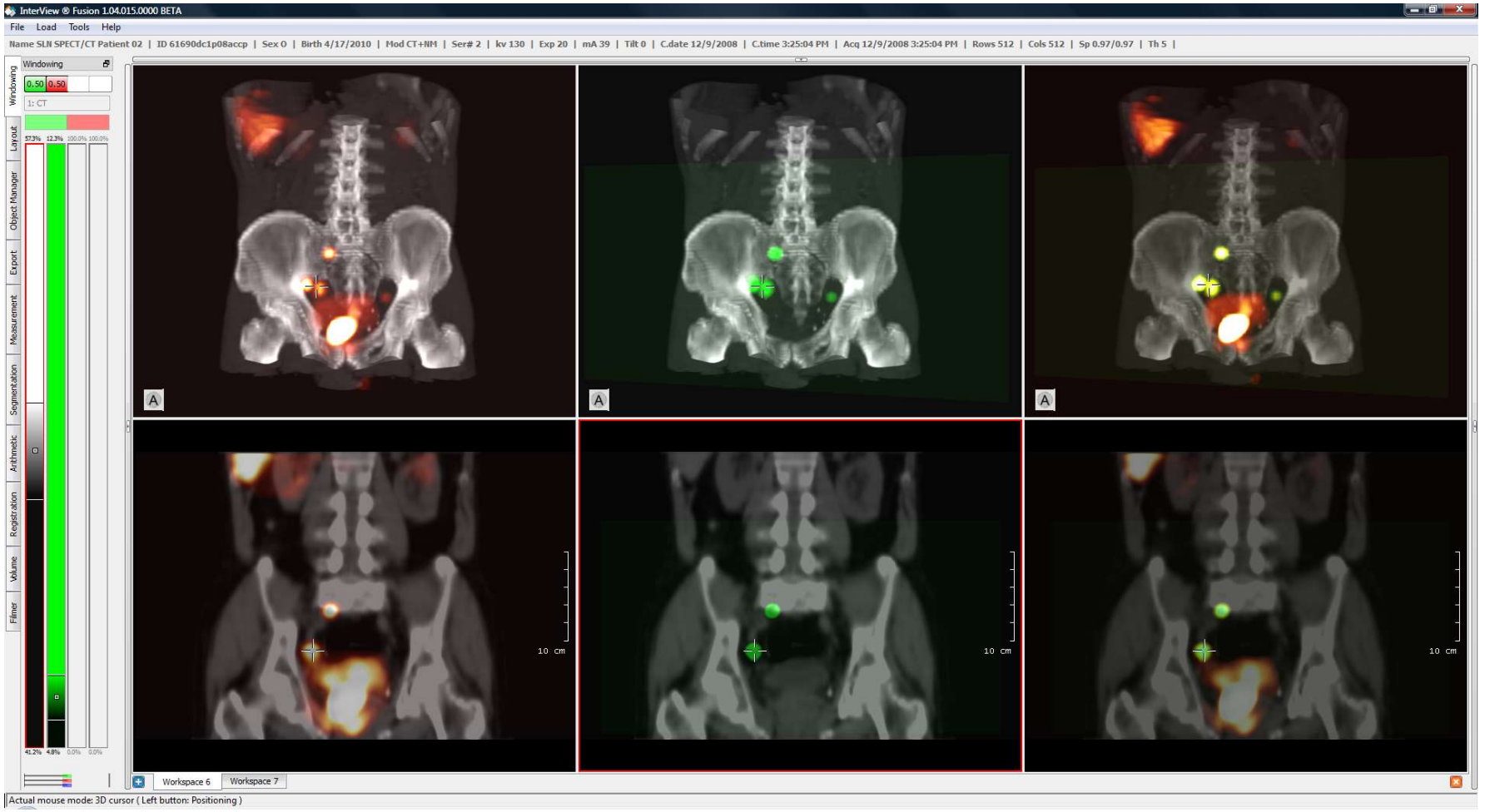
# Segmentation

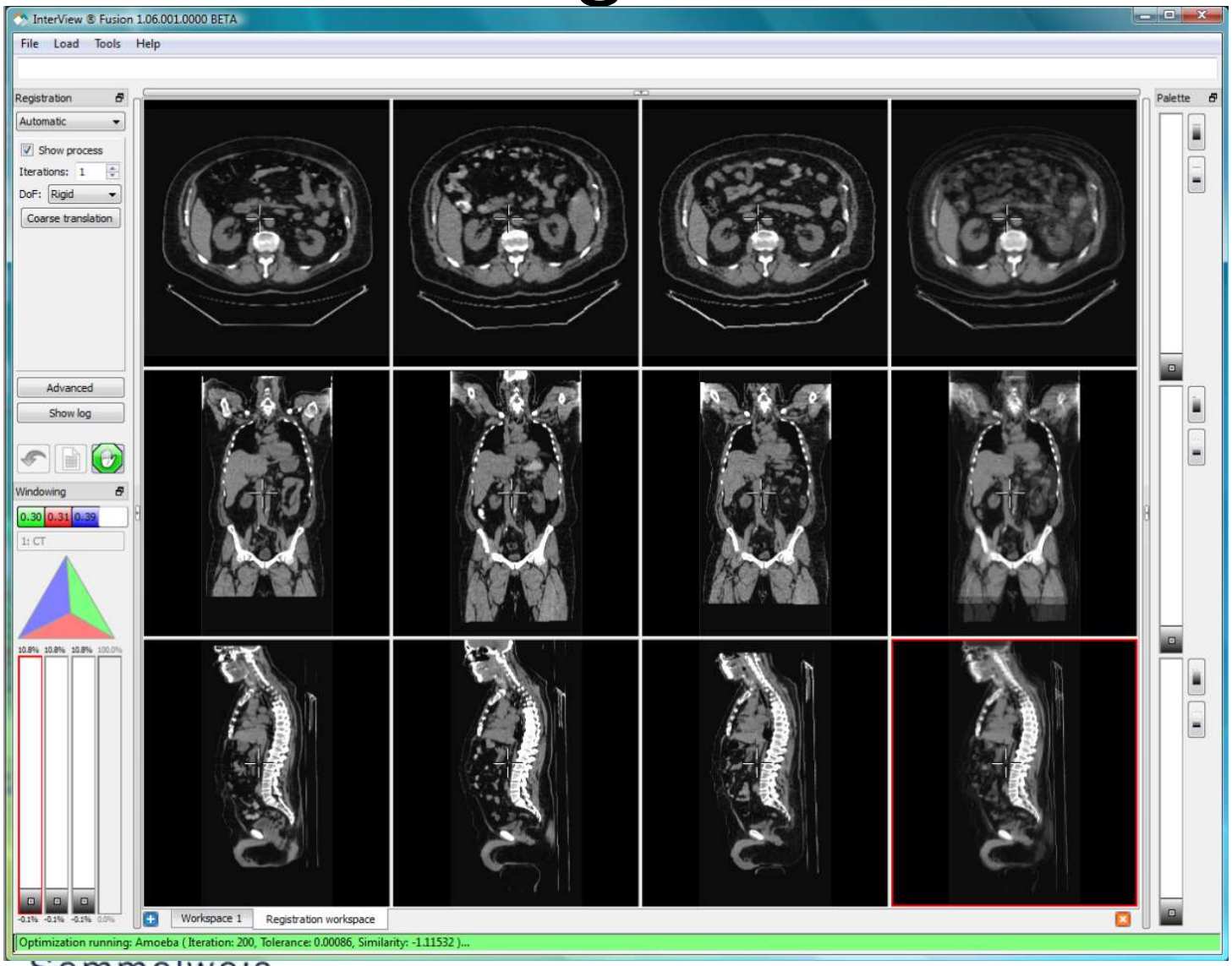
SLN SPECT-CT

Segmented SPECT-CT

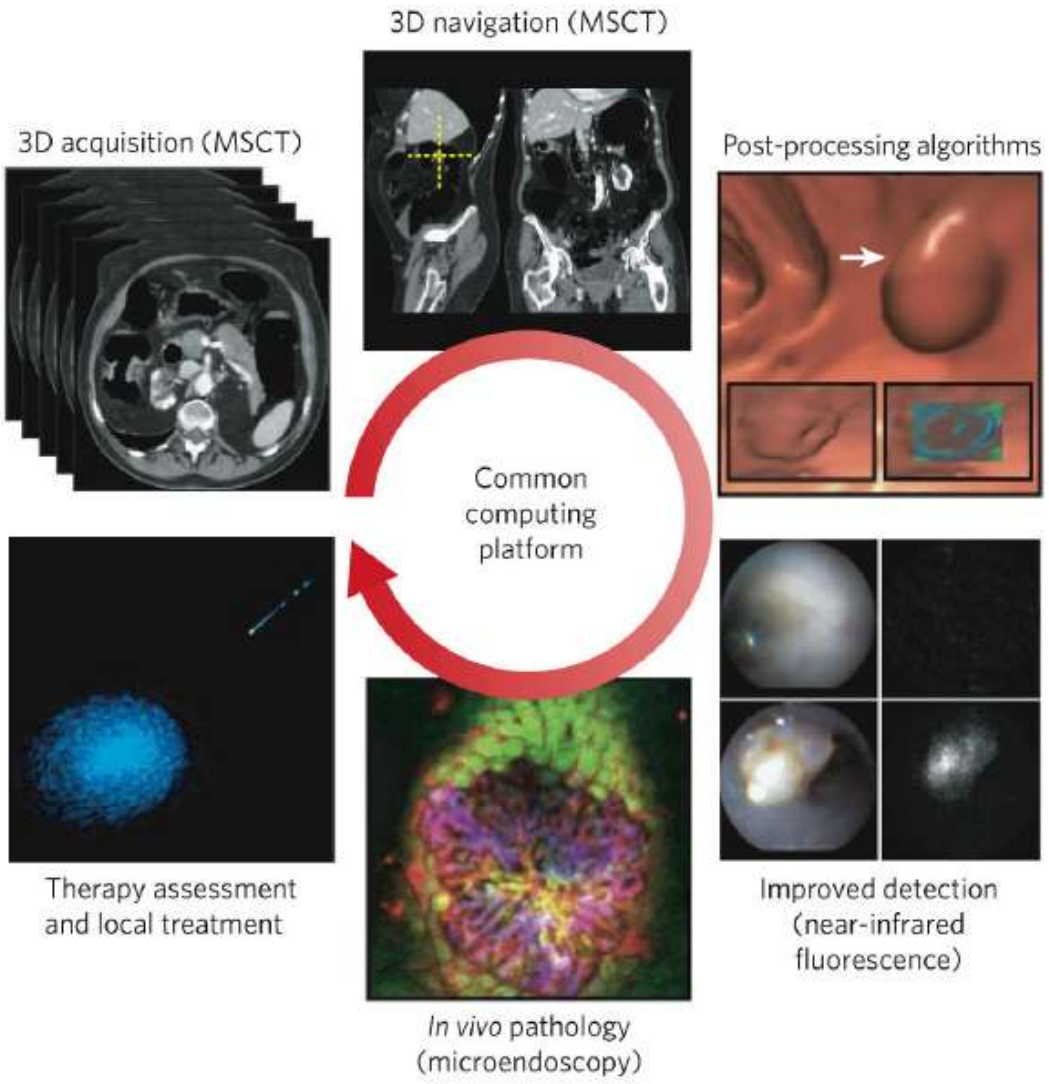
Triple fusion (Segmented SPECT-SPECT-CT)



# Co-registration



# Imaging in Clinical PACS (MGH)



# Thanks!

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