

# MOLECULAR IMAGING

Functional Imaging Methods  
Multi-Modality



Máthé, Domokos PhD

Nanobiotechnology and In Vivo Imaging Center

## 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



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- 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.”

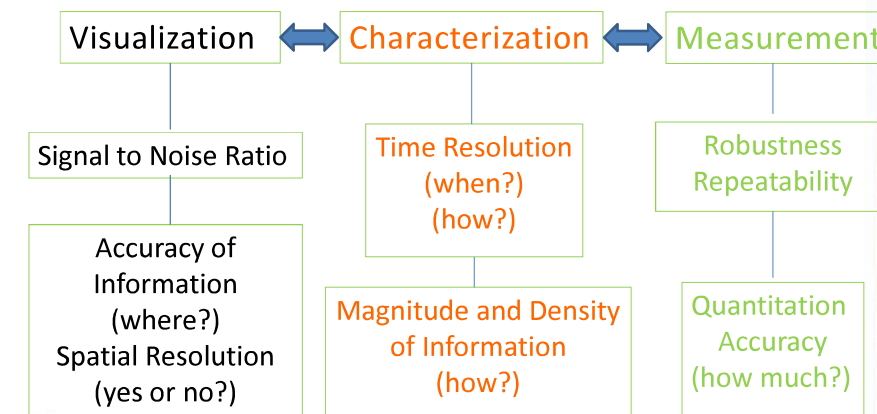
(definition by the U.S. Society of Nuclear Medicine and Molecular Imaging)



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## Molecular Imaging Ideals:

**B i o l o g i c a l**

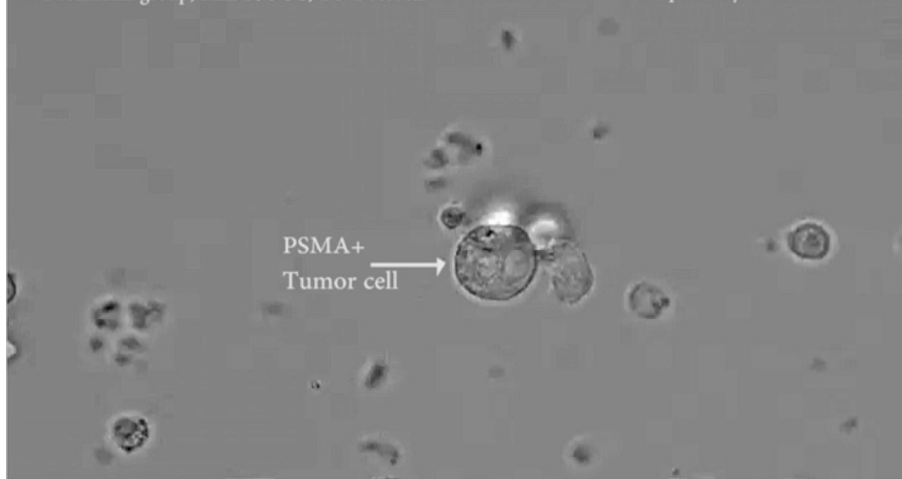


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## Tumor cell killing with T-cells targeted against PSMA molecule using a Targeting Bridge Module (TM)

© Bachmann group, HZDR/UCC, TU-Dresden

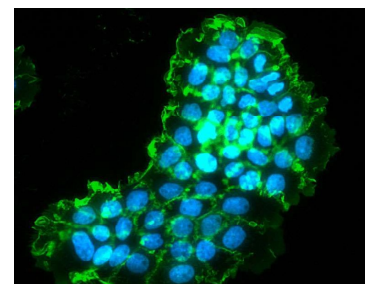
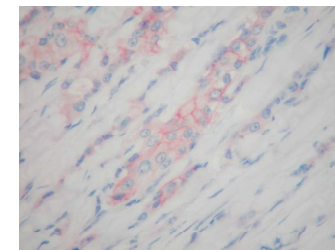
Prepared by: Nicola Mitwasi



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## Microscopy: Immunohistochemistry, immunofluorescence

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



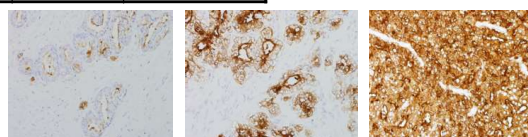
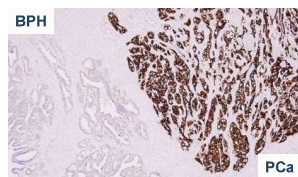
A431 epithelial cc. tumor cells, nuclei stained with Hoechst-blue, PHOSPHORYLATED EGF receptors in cell membrane are GREEN using an antibody coupled to 488 nm emission DyLight fluorescent stain



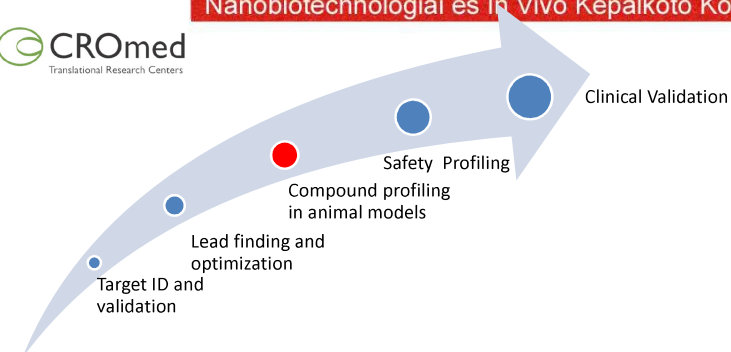
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## PSMA Expression is Prostate Cancer Specific and Increases with Tumor Grade

# Cases Studied	% Cases Reported to be PSMA Positive	Reference
251	94%	Wright et al
184	100%	Bostwick et al
51	84%	Mannweiler et al
42	88%	Kusumi et al
21	100%	Ananias et al
905	99.9%	Loda et al



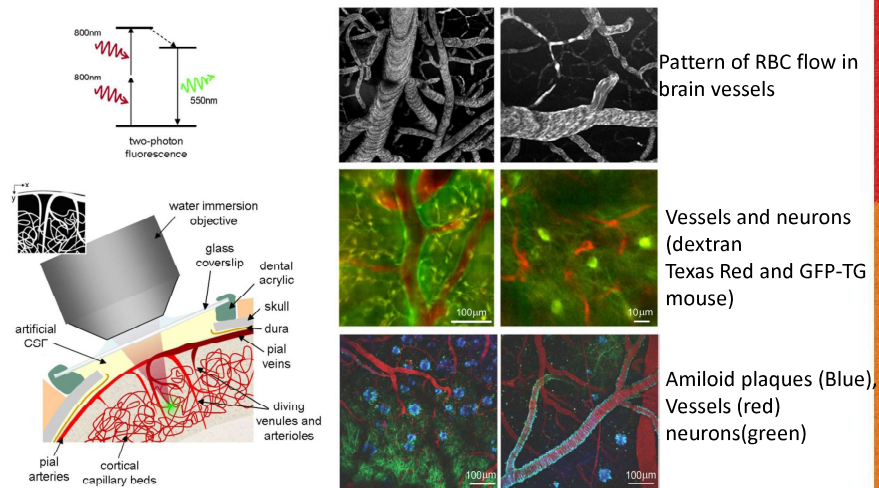
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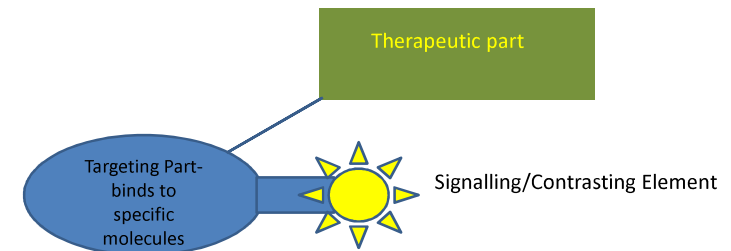
DRUG	Indication	Probe	Modality
Vortioxetine	Major depressive disorder	<sup>11</sup> C-MADAM <sup>11</sup> C-WAY100635	PET
Topiramate	Epilepsy	Blood oxygenation/flow	fMRI
Imatinib	GI stromal tumors	<sup>18</sup> F-FDG; composition; density	PET/CT; CT; MRI
Rotigotine	Parkinson's Disease	<sup>99m</sup> Tc-TRODAT-1	SPECT
Pazopanib	Soft tissue sarcoma	<sup>99m</sup> Tc-VEGF; Perfusion	SPECT; DCE-MRI
Pertuzumab	Breast cancer	<sup>99m</sup> Tc-2Rs15d; <sup>18</sup> F-FDG	PET/CT; SPECT/CT

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

better S/N ratio  
increased resolution



# General Structure of a Molecular Probe



Small Molecules  
Peptides  
Proteins/Subunits  
Antibodies/Subunits

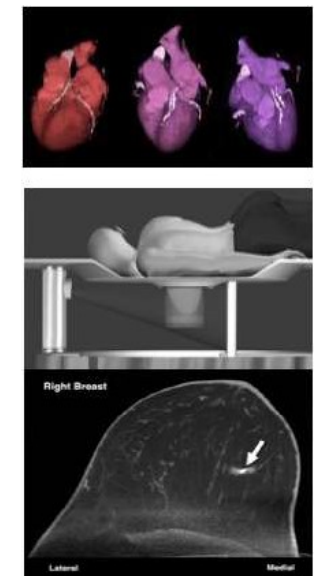
## Chemical Nature:

PET/SPECT: isotopes (radioactivity)  
Optical/Acoustic: Fluorescent Dyes  
MRI: Gd, Fe  
CT: iodine, Barium sulphate  
Nano/microparticles:  
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 Good time resolution Percs kép-idők Költségesen drága Anatómiai módszer	Sugárterhelés Rossz lágyrészt kontraszt Jelenleg csak anatómiai és funkcionális képalk.	Ba, I, Kr, Xe	Tumor perfúzió,
PET	Bármilyen mélységű kép Egésztest-képkalkítás Kvantitatív mérések Kombinálható CT/MRI-vel	Sugárterhelés Drága Milliméteres felbontás 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 Egésztest-képkalkítás Kvantitatív mérések Multiplex Teragnosztika Kombinálható CT-vel	Sugárterhelés Szubmilliméteres-milliméteres felbontás Hosszabb képidők	Tc-99m, I-123, In-111, Lu-177	Molekuláris diagnosztika Radioterápia (NHL, NET, pm. cc.)
MRI	Bármilyen mélységű kép Egésztest-képkalkítás Nincs ionizáló sugárzás Kitudó lágyrészt kontraszt	Drága Hosszú képidők Korlátozott érzékenység	Gd <sup>3+</sup> , vas-oxid részecskék (SPIO, USPIO)	Prostata daganat nycs. met. Fokális májleziók Szív perfúzió
MRS	Nincs ionizáló sugárzás Egésztest-képkalkítás	Drága Hosszú képidők Kis érzékenység	Kolin, laktát, kreatin, lipidek, N-acetil-aszpartát	Agytumorok anyagcsereje Alzheimer-kór követése
UH	Nincs ionizáló sugárzás Rövid/valós idejű képkalkítás Nagy térbeli felbontás Olcsóság Nagy érzékenység	Egésztest-képkalkítás nincs Kontrasztanyagok csak az érrendszerre Operátor-függő	Mikro-buborékok	Fokális májleziók, echokardiográfia, Tumor perfúzió
Optikai módszerek	Nincs ionizáló sugárzás Rövid/valós idejű képkalkítás Nagy térbeli felbontás Olcsóság Nagy érzékenység, kvantitatív Multiplex	Korlátozott áthatolóképesség (1 cm) Nincs egésztest-képkalkítás	Fluoreszcens molekulák és festékek, fény-elnyelő nanorészecskék	OCT-érlemezsedés, retinopathiák, kolonoszkópia

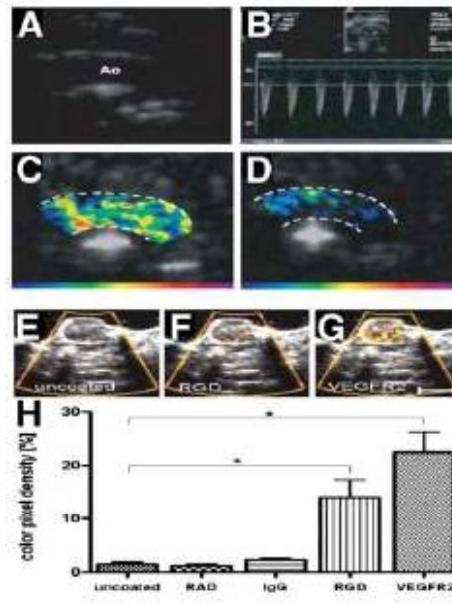
CT	3D Attenuation Map of X-Rays
Advantages	Any Imaging Depth Good Resolution Simple Medium-Priced Sub-minute scan times
Disadvantages	Radiation Dose Soft Tissue C. Just anatomical and functional images Any imaging depth Good Resolution WB Imaging Minute Scanning Time Semi-Expensive Anatomy
Contrast Materials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





## Nanobiotechnológiai és In Vivo Képalkotó Központ

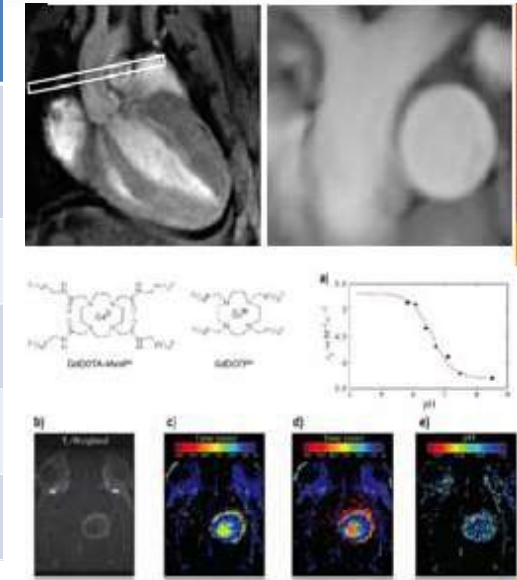
UltraSound	3D differences in echo and spreading of sound waves
Advantages	No ionizing radiation Fast/RT imaging High Sensitivity High Resolution Cheap
Disadvantages	No WB img. Only vascular contrast materials Operator dependency
Contrast Materials	Micro-Bubbles
Clinical Use	Focal liver lesions, Echocardiography,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



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## Nanobiotechnológiai és In Vivo Képalkotó Központ

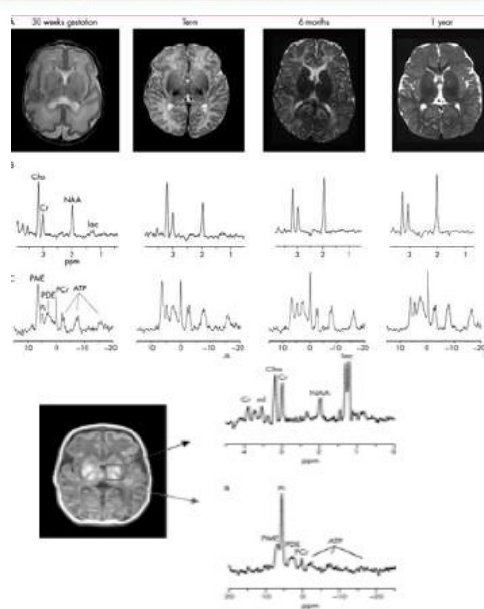
MRI (IRM...)	3D / 2D Image of Proton Spins
Advantages	Any Imaging Depth and Plane WB Imaging No ionizing radiation High Soft Tissue Contrast
Disadvantages	Expensive Low Sensitivity Long imaging time
Contrast Materials	Gd3+, iron-oxide particles (SPIO, USPIO)
Clinical Use	Liver, Brain Lesions, Cardio-MRI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



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## Nanobiotechnológiai és In Vivo Képalkotó Központ

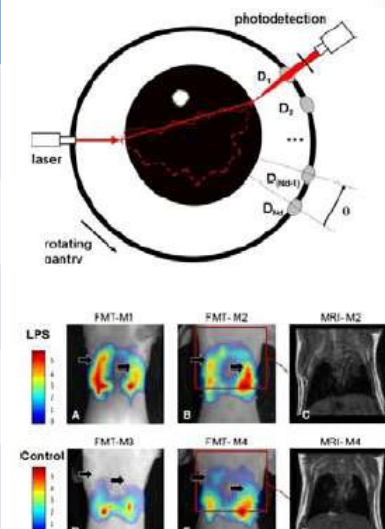
MRS	Recorded Electromagnetic Resonance Spectrum
Advantages	No ionizing radiation WB Imaging
Disadvantages	Expensive Very low sensitivity External Calibration / Special Knowledge
Contrast Materials	Cholin, Lactate, Creatin, Lipids, N-Ac-Aspartate
Clinical Use	Brain Tumor Stratification, Stroke
Voxel Sizes,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



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## Nanobiotechnológiai és In Vivo Képalkotó Központ

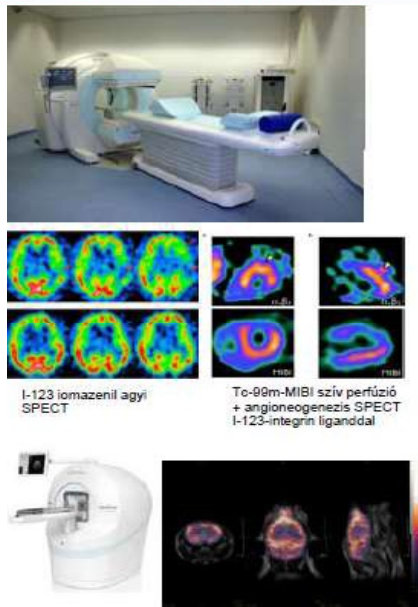
Optical Methods	Visible/NIR Light Transmission/Reflectance/Emission/ Scatter in 2D, 3D
Advantages	No ionizing radiation Short/RT Imaging High Spatial Resolution Very Sensitive, Semi-Quantitative Multiplex
Disadvantages	Limited Transparency No WB imaging
Contrast Materials	Fluorescent molecules, Light-emitting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



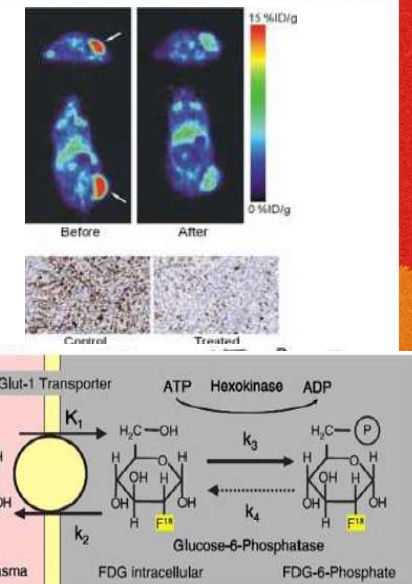
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SPECT	3D Gamma-ray Source Array in Tissue
Advantages	Any imaging depth WB Imaging Quantitative Good resolution Multiplexing Theranostics Combination w/CT
Disadvantages	Radiation Dose Sub-mm Resol. Long imaging times
Contrast Materials	Tc-99m, I-123, 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 x 0.8 mm Small A: 0.3 x 0.3 x 0.3 mm 2 pM / voxel 1/10 atom per cell



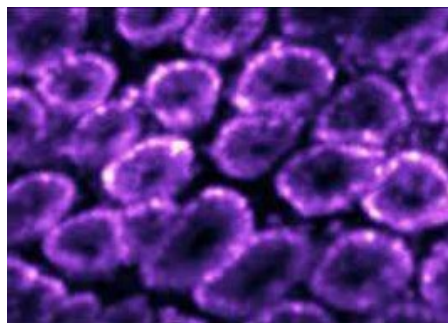
PET	3D 511 keV Gamma-ray Source Array in Tissue
Advantages	Any imaging depth WB Imaging Quantitative Anatomical Co-registration CT, MRI
Disadvantages	Radiation Dose PRICE Resolution Longer imaging times
Contrast Materials	C-11, F-18, Ga-68, Cu-64, Zr-89
Clinical Use	FDG Patient Staging+Follow-
Voxel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



## Screening-Confocal Endomicroscopy



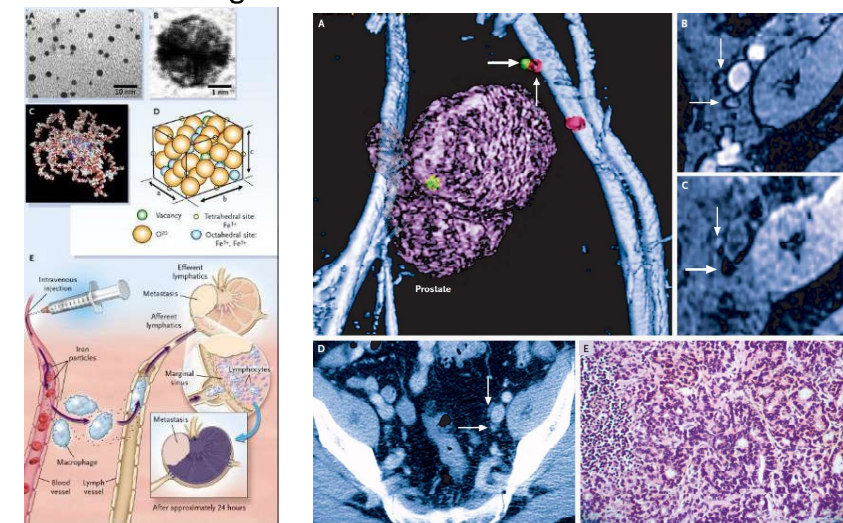
Oesophagus, stomach, bile ducts, ileal/colonic mucosa examined on cell level using fiber optic confocal microscope at autofluorescence emission/excitation wl-s



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



## Diagnostics-USPIO MRI-A SAD STORY



Harisingani M et al N.Engl. J.Med. 2003





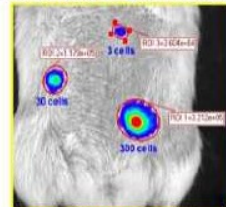
**Bioluminescent Imaging:** Only external genes built-in to germlines (whole animal) or plasmids for reporter gene imaging.

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

Luciferine enzyme production  
Precision filters  
Low f-number and large diameter lens

Gives high sensitivity and uniform light collection

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)

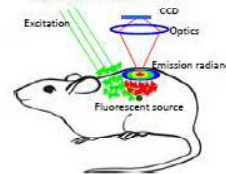


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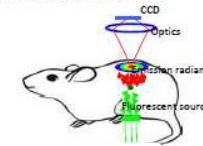
FLUORESCENCE IMAGING

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.

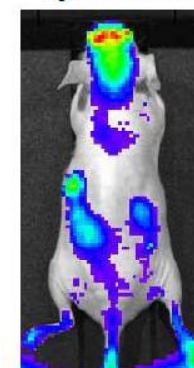
Reflectance



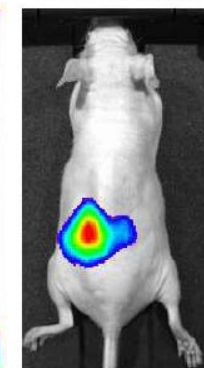
Transmission



Epi-Illumination



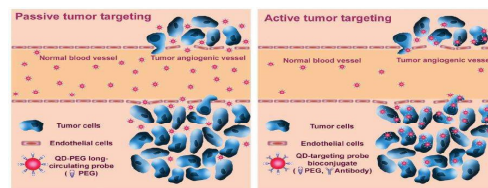
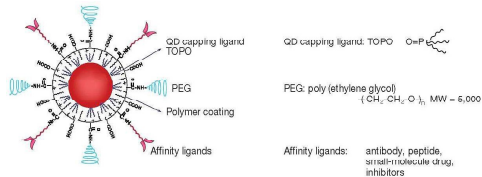
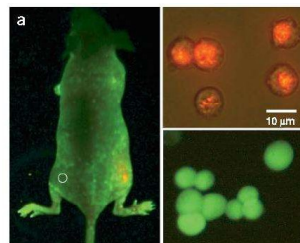
Transillumination



Pillow implanted medial to left kidney, 1x1015 molecules

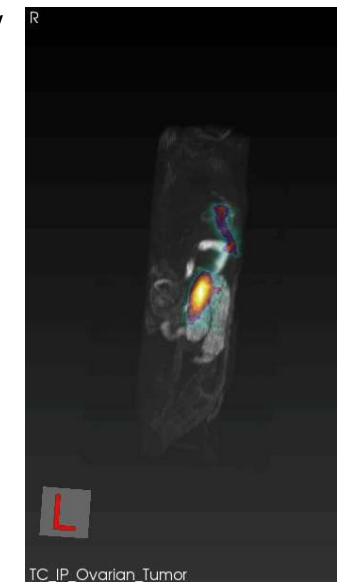
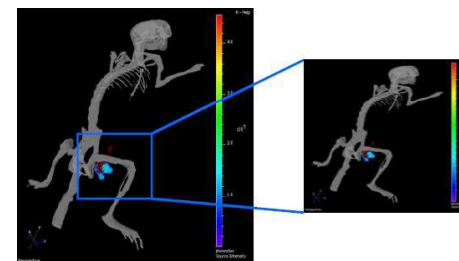


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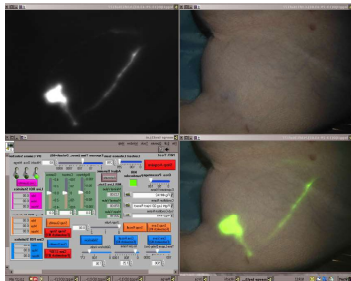
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3D Optical Tomography



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## Image guided surgery

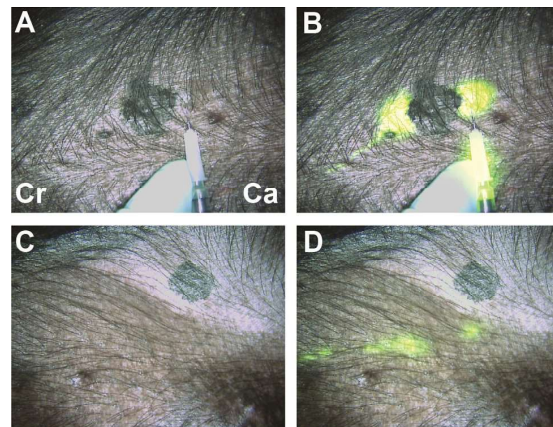


John Frangioni FLARE Harvard



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## Planar Fluorescence In the Clinic

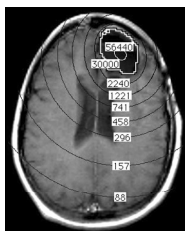
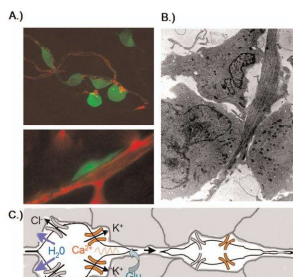
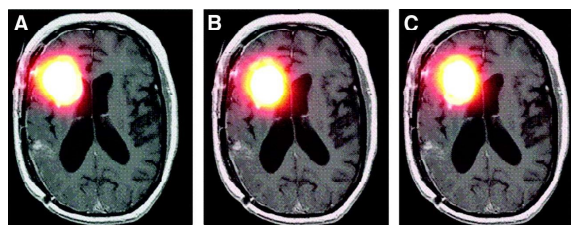


Melanoma, ICG-Albumine particle



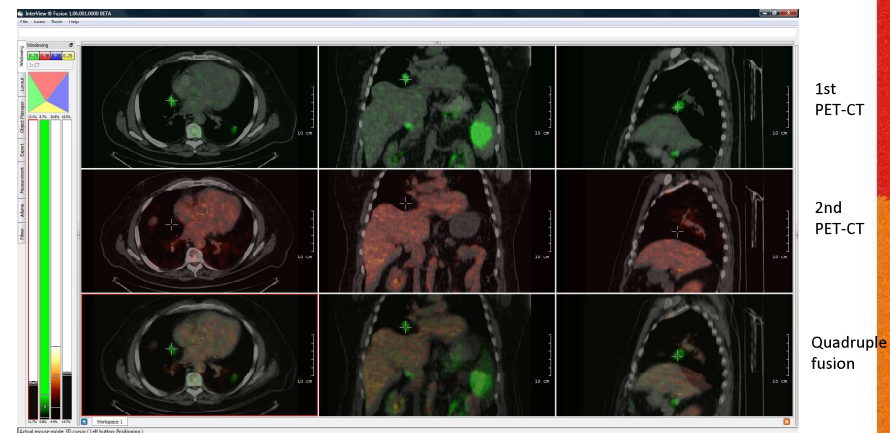
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## Personalized Targeted Therapy: SPECT/MRI/RNT



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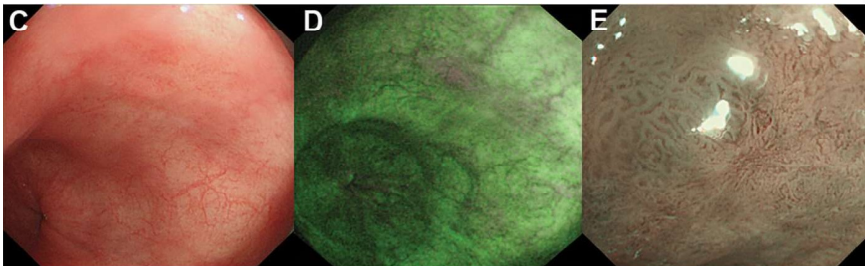
## Monitoring with FDG-PET



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## Planáris autofluoreszcencia-endoszkópia



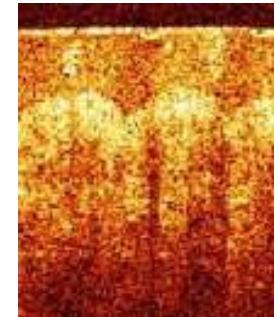
C: Barrett-oesophagus endoszkópia, D: autofluoreszcencia (lila) E: nagyfelbontású endoszkópia-irreguláris mintázat. A biopszia adenocc.-t igazolt.



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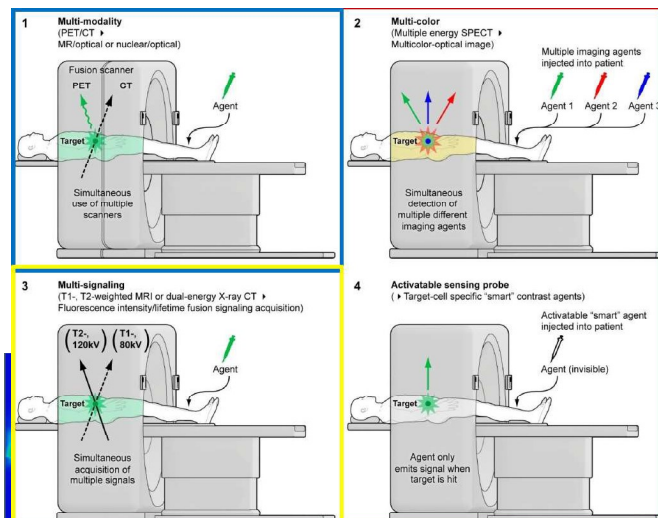
## OCT (optical coherence tomography)

- Retina:
- Mucosák
- Porc
- Agyi vérkeringés
- Pár mm-es áthatolóképesség
- Mikronos felbontás
- Valós idejű képalkotás



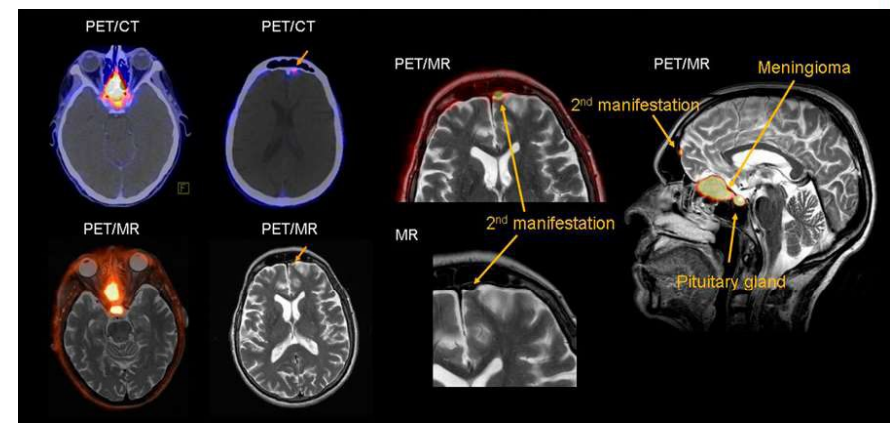
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## Multimodális rendszerek lehetőségei



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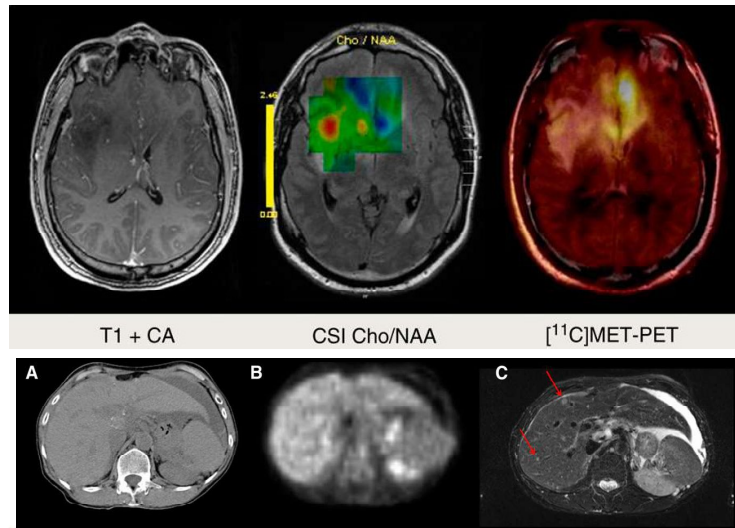
## PET/MRI



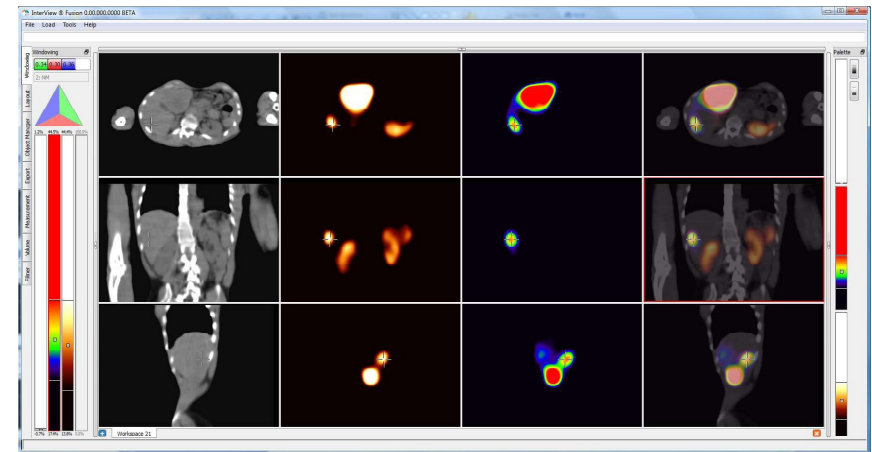
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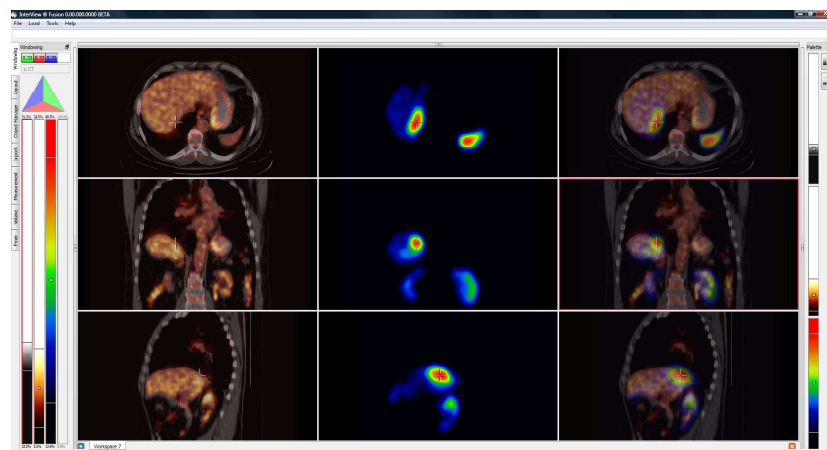
## PET/MRI



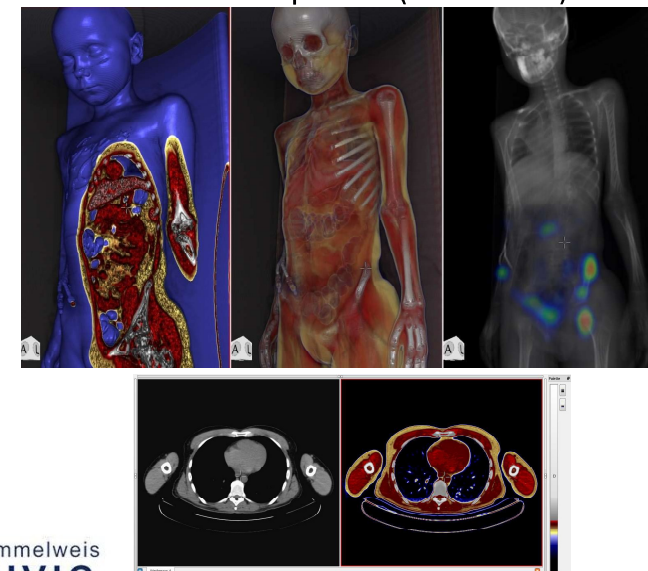
SPECT for neuro-endocrine tumor  
treatment/assessment



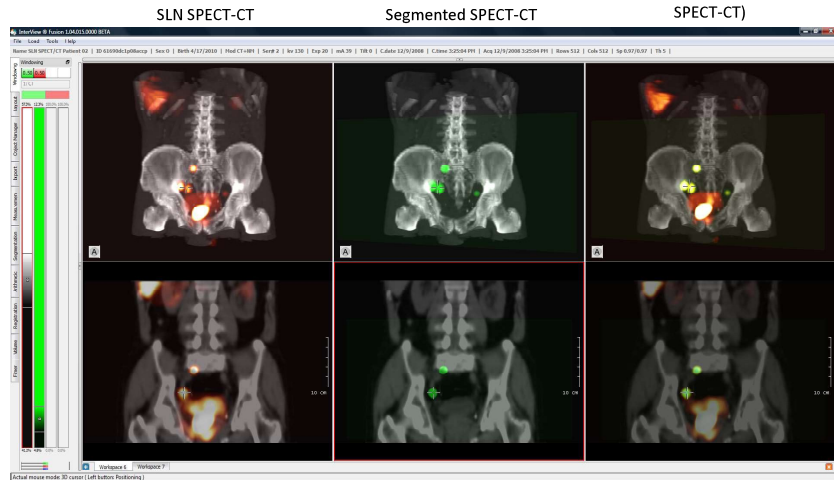
## PET/SPECT/CT – Funkcionális és morfológiai információ együtt



SPECT-CT look-up table (color scale)

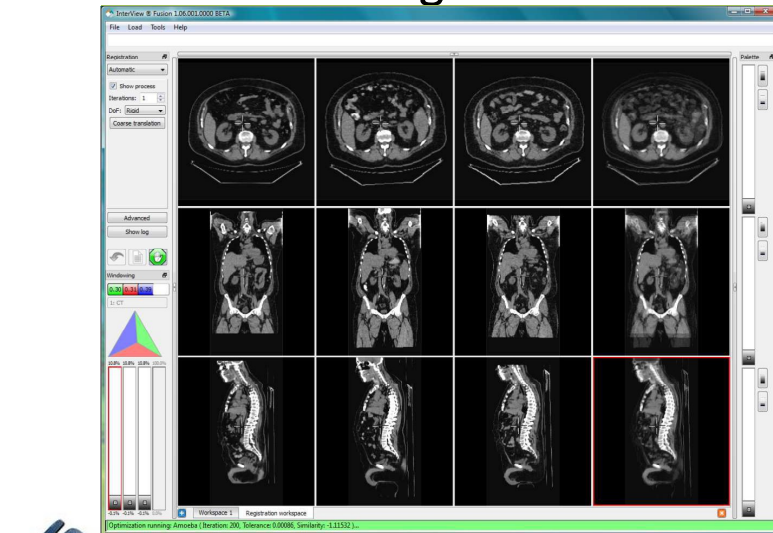


## Segmentation



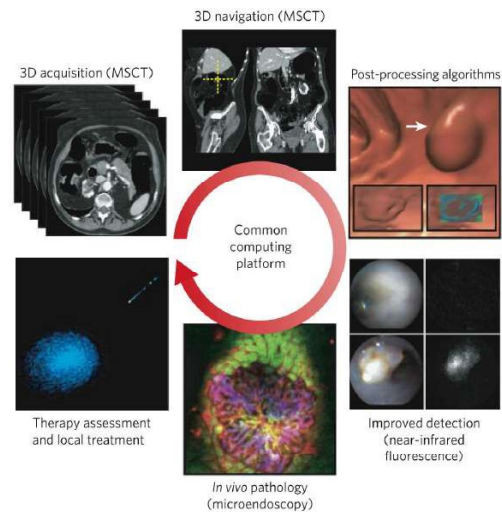
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## Co-registration



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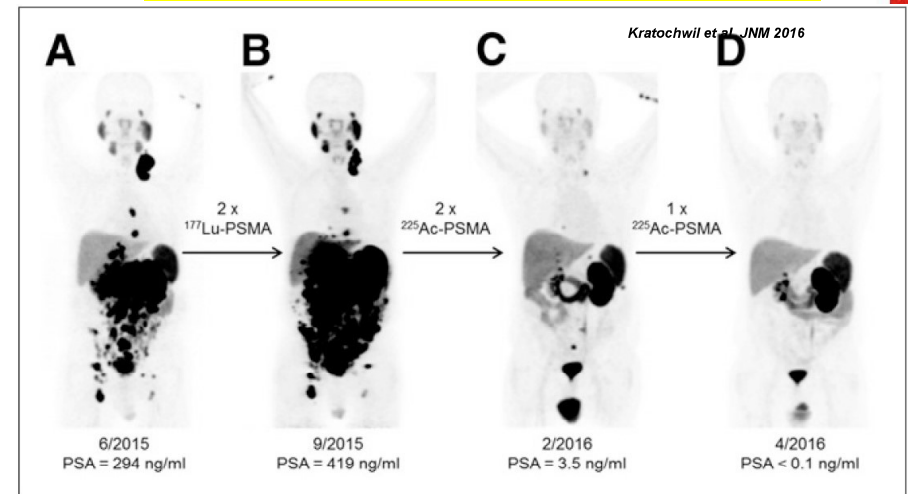
## Imaging in Clinical PACS (MGH)



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Alpha radionuclide Therapy using Actinium-225 labeled PSMA ligand small molecule

**Curing the presently incurable...**



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Thanks!

- [domokos.mathe@cromedresearch.com](mailto:domokos.mathe@cromedresearch.com)

