Metabolic Seminar – 23rd of January 2013

Institute of Physiology AS ČR, p.r.i.

μPET/CT

Molecular imaging in small laboratory animals

Nuclear Physics Institute AS ČR, p.r.i.

Sebastian Eigner, ORF, NPI AS ČR, p.r.i.

Molecular Imaging

In vivo molecular imaging in small animals is the bridge between *in vitro* data and translation to clinical application

Multiple longitudinal images provide more reliable information and reduce animal numbers

Discover new predictive imaging biomarkers

Accelerate the pre-clinical validation of new drugs

Enable selection of drug candidates for clinical translation

No one imaging method is ideal for all studies

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Morphological Imaging

CT

WANTED

Lowly Wurm

Out of: Richard Scarry´s Children´s Books

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Functional Imaging

PET

WANTED

Lowly Wurm

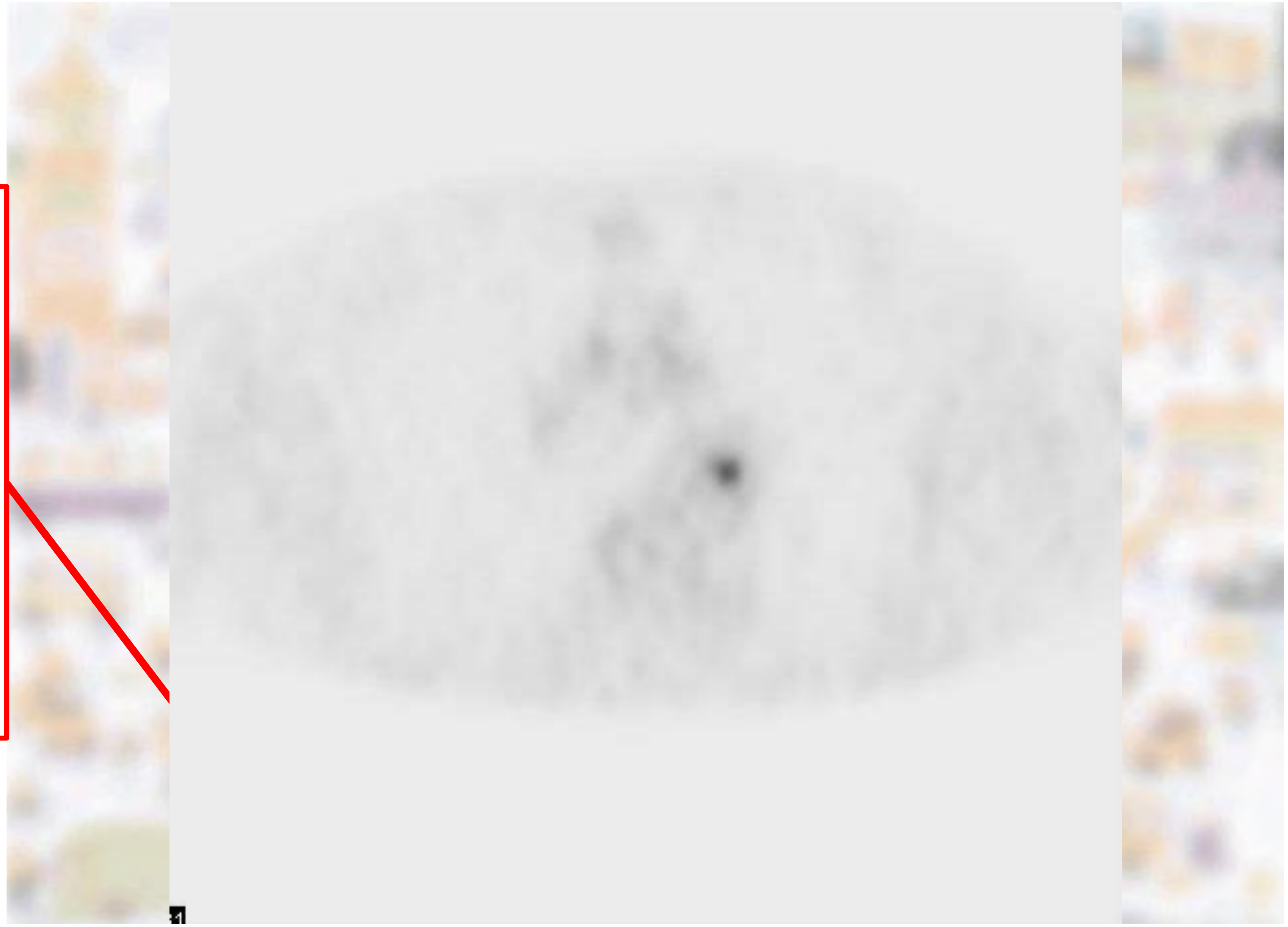

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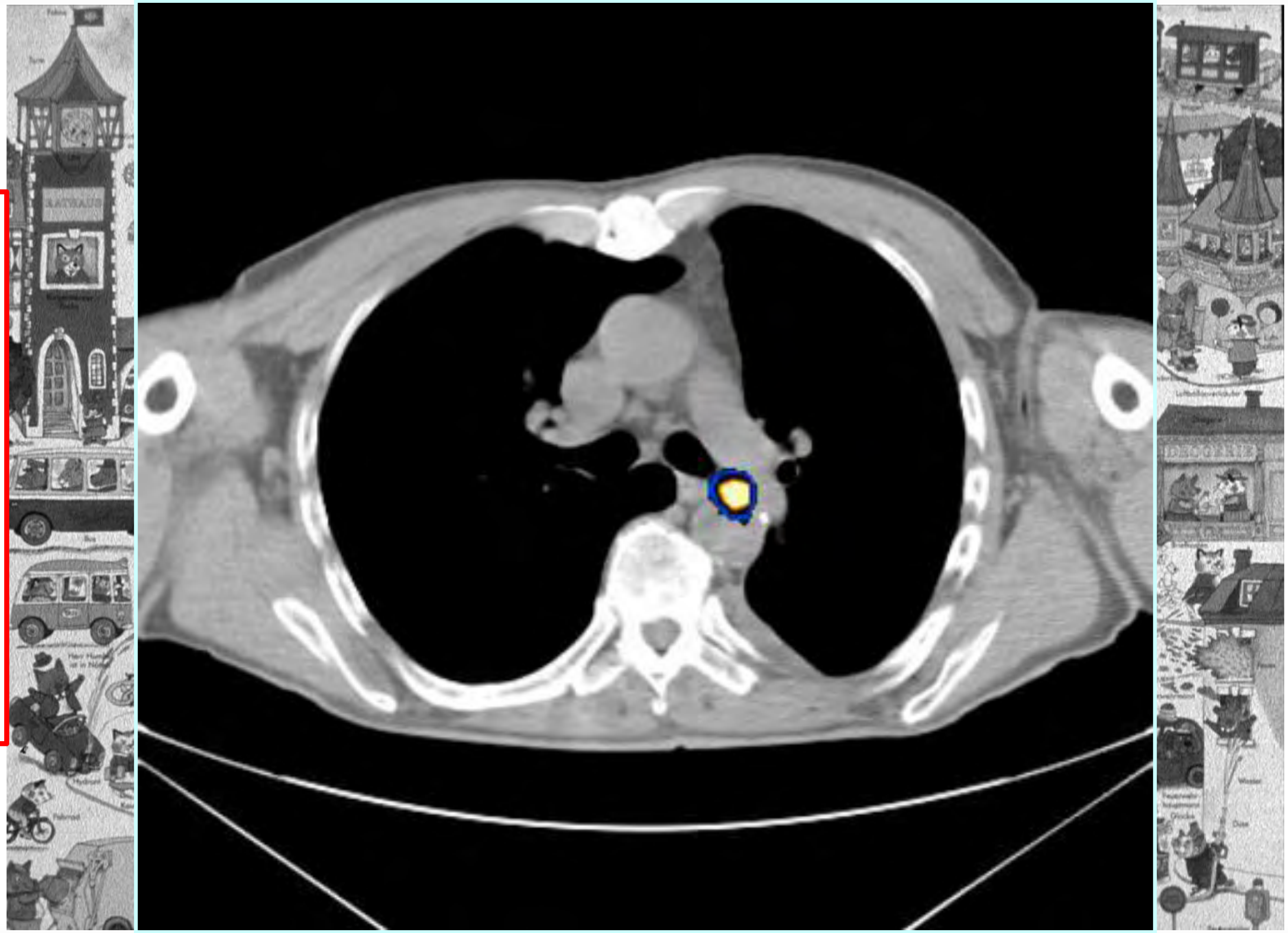
Image Fusion

PET/CT

WANTED

Lowly Wurm



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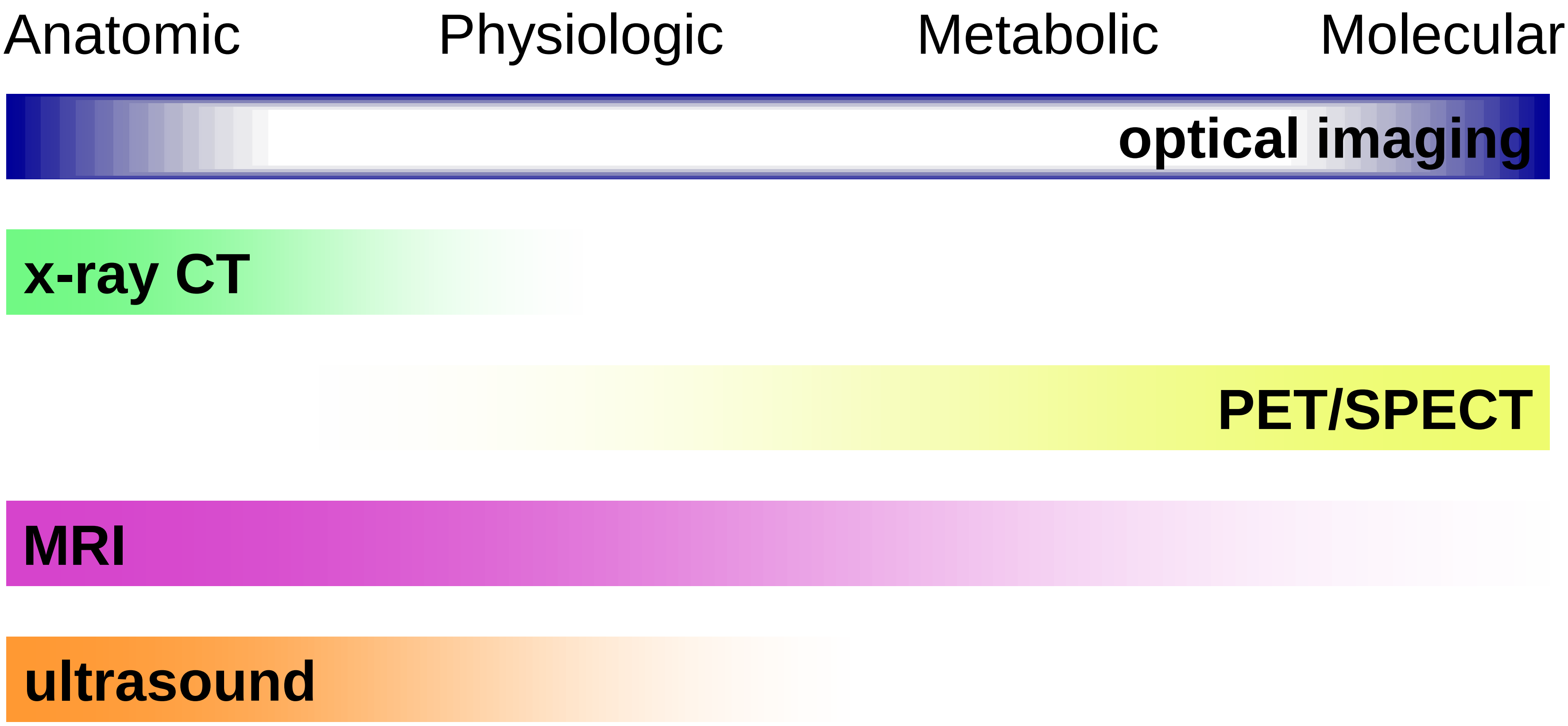
In vivo biomedical imaging technologies



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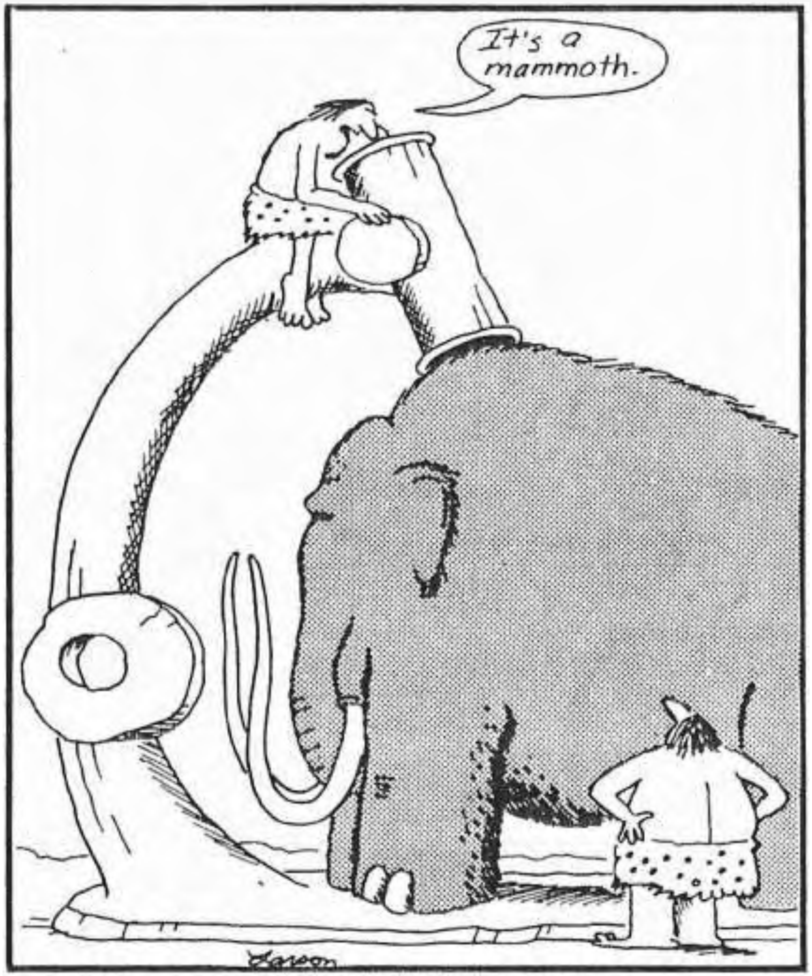
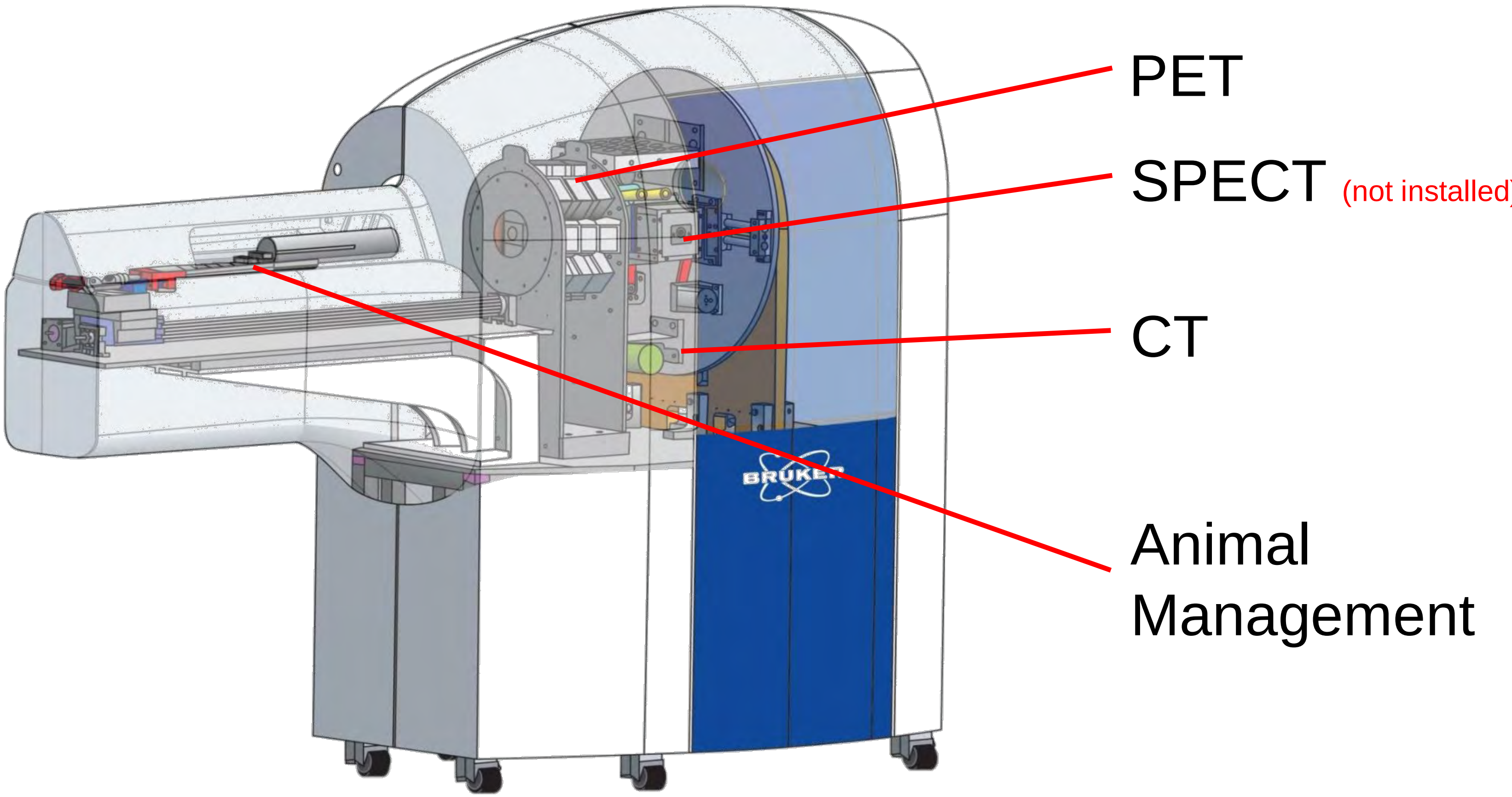
Molecular Imaging: The Big Picture



Early Microscope

Albira

PET/SPECT/CT



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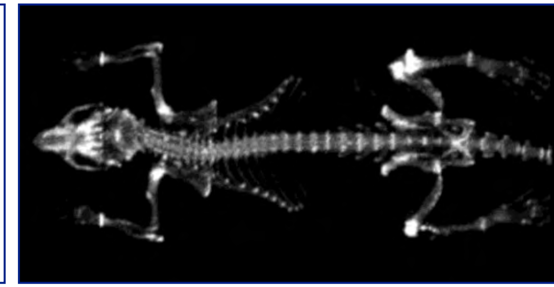
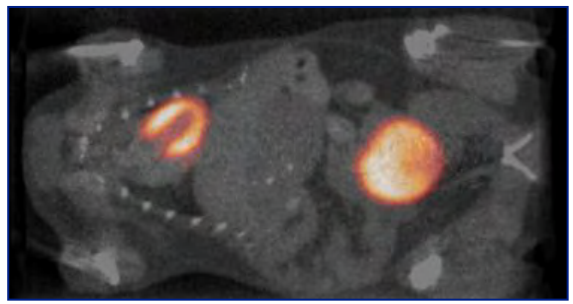
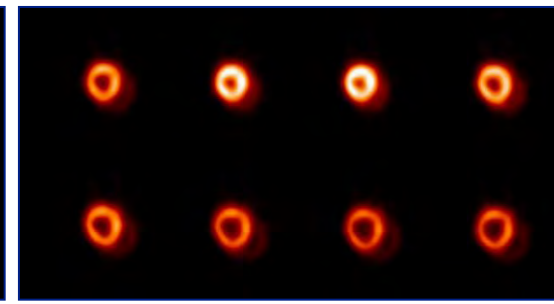
Small Animal Imaging

Requirements

High spatial resolution

- mouse organs ~1000-fold smaller volume than human

High sensitivity

- number of targets also smaller, radiation dosimetry can be limiting

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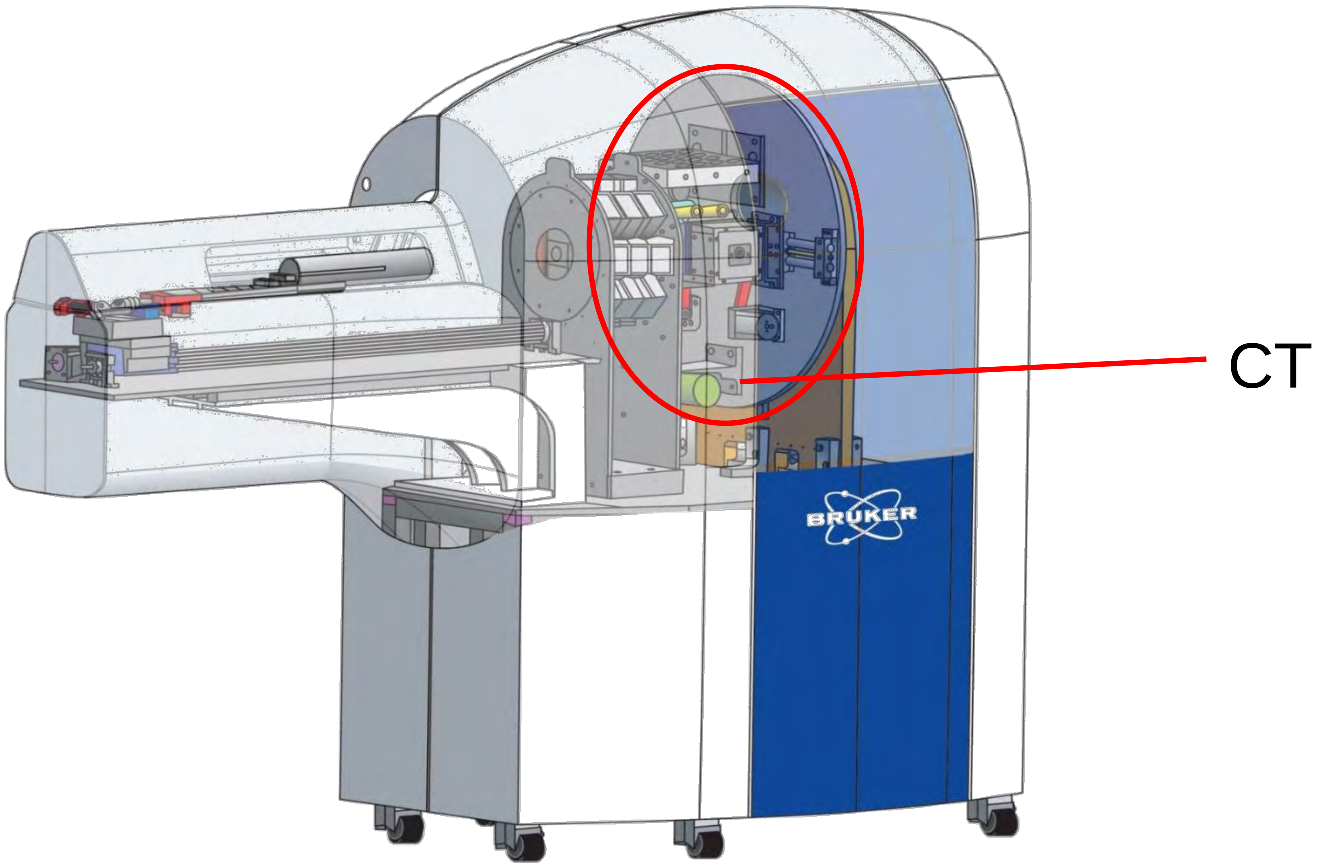
Computed X-ray tomography

CT



Computed X-ray tomography

CT

ADVANTAGES

- high spatial resolution (20µm)
- easy to operate
- relatively cheap

DISADVANTAGES

- sensitivity = milli-molar
- No functional information
- High radiation source
- Bad soft tissue contrast
- Contrast agents required to improve soft tissue contrast

CT is based on the measurement on X-ray attenuation (~ 40-80 keVp)

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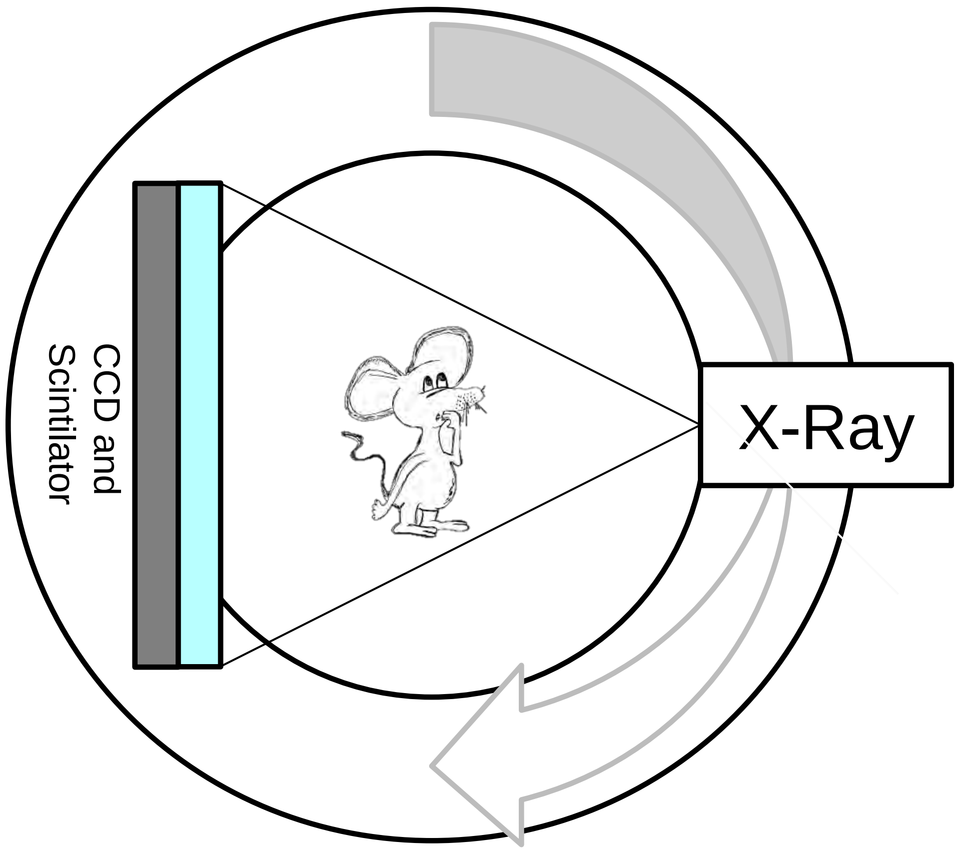
Computed X-ray tomography

Tomography Principle

X-ray source and X-ray-detector are mounted on a common rotational stage



Projection data are acquired in a **step-and-shoot (SAS) mode**

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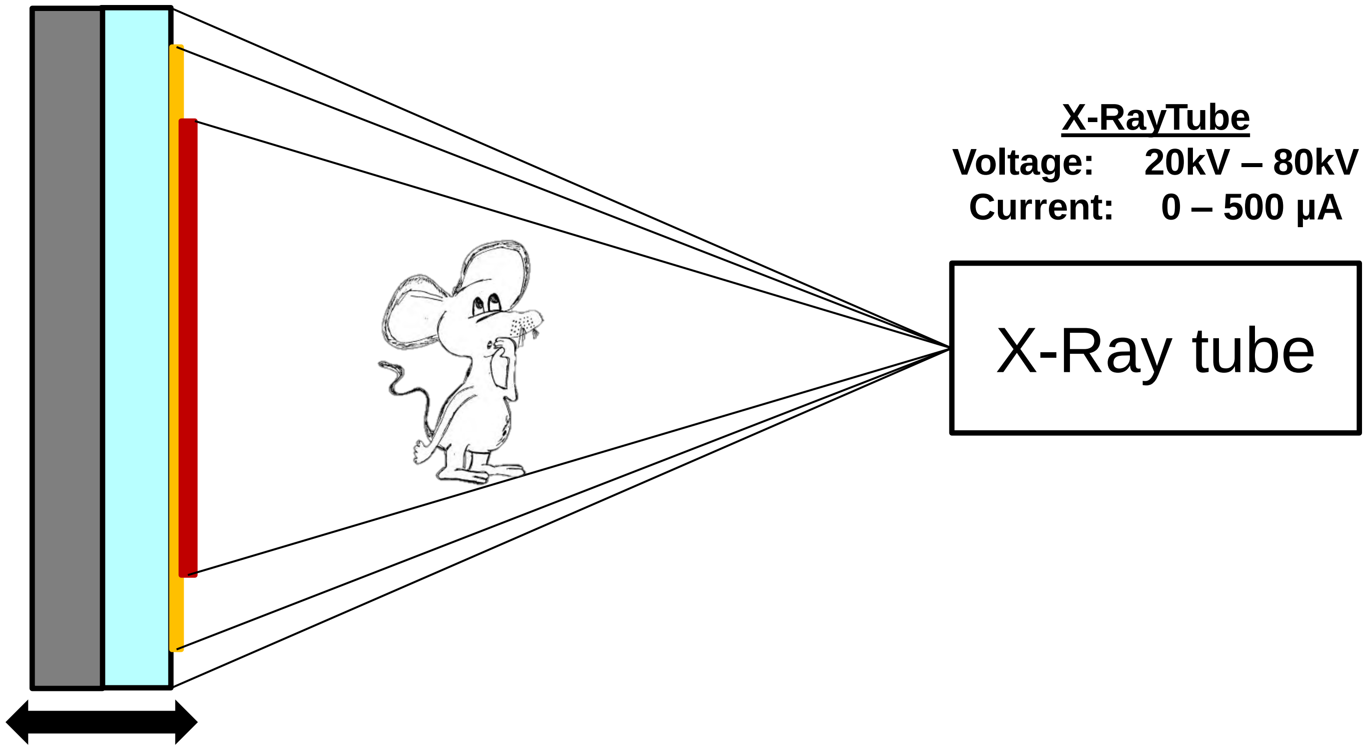
Computed X-ray tomography

CT-Detector Principle

CCD-Array and **Scintilator**
Scintilator: NaI
CCD Array: Large Area (2048 x 3072, 33microns pitch)



Move detectorpanel to adjust FOV and resolution

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Computed X-ray tomography

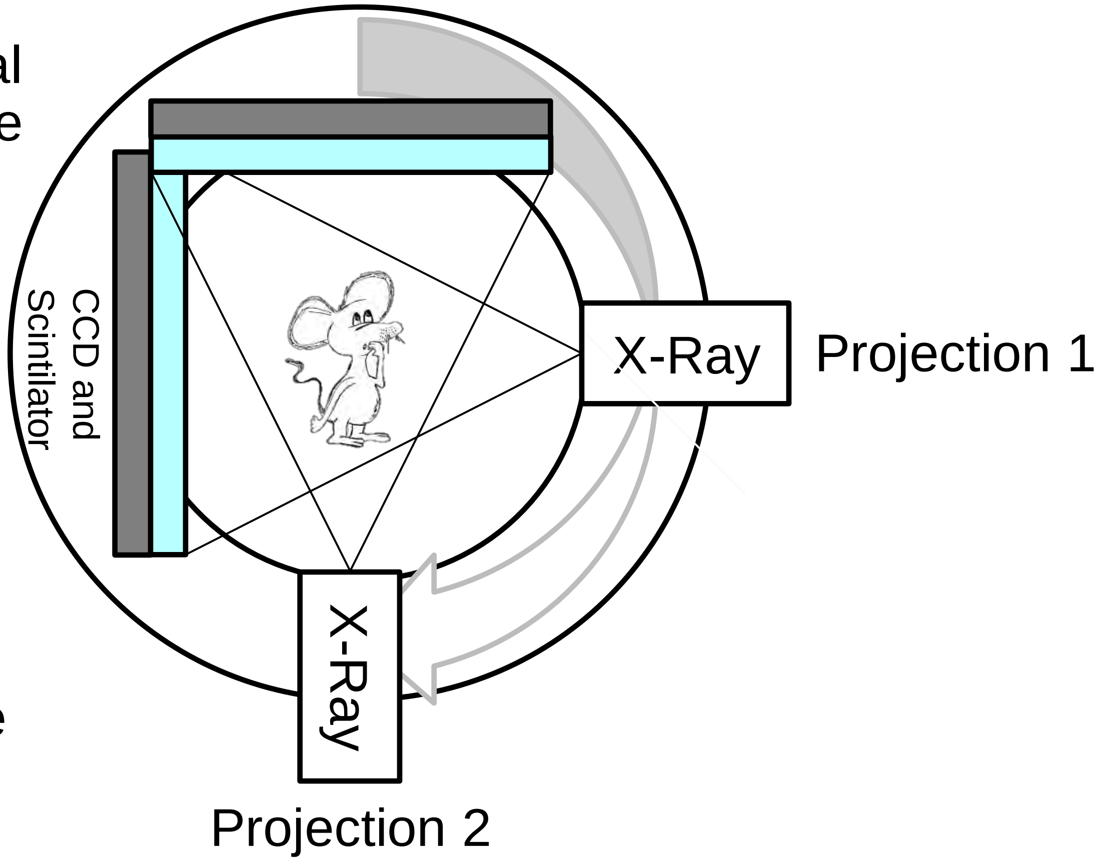
Step-and-Shoot (SAS) Mode

N projections over an total rotation angel of 360° are acquired

N = number of steps

Rotation angle per step depends on preselected resolution

The higher the reolution , the more steps are required



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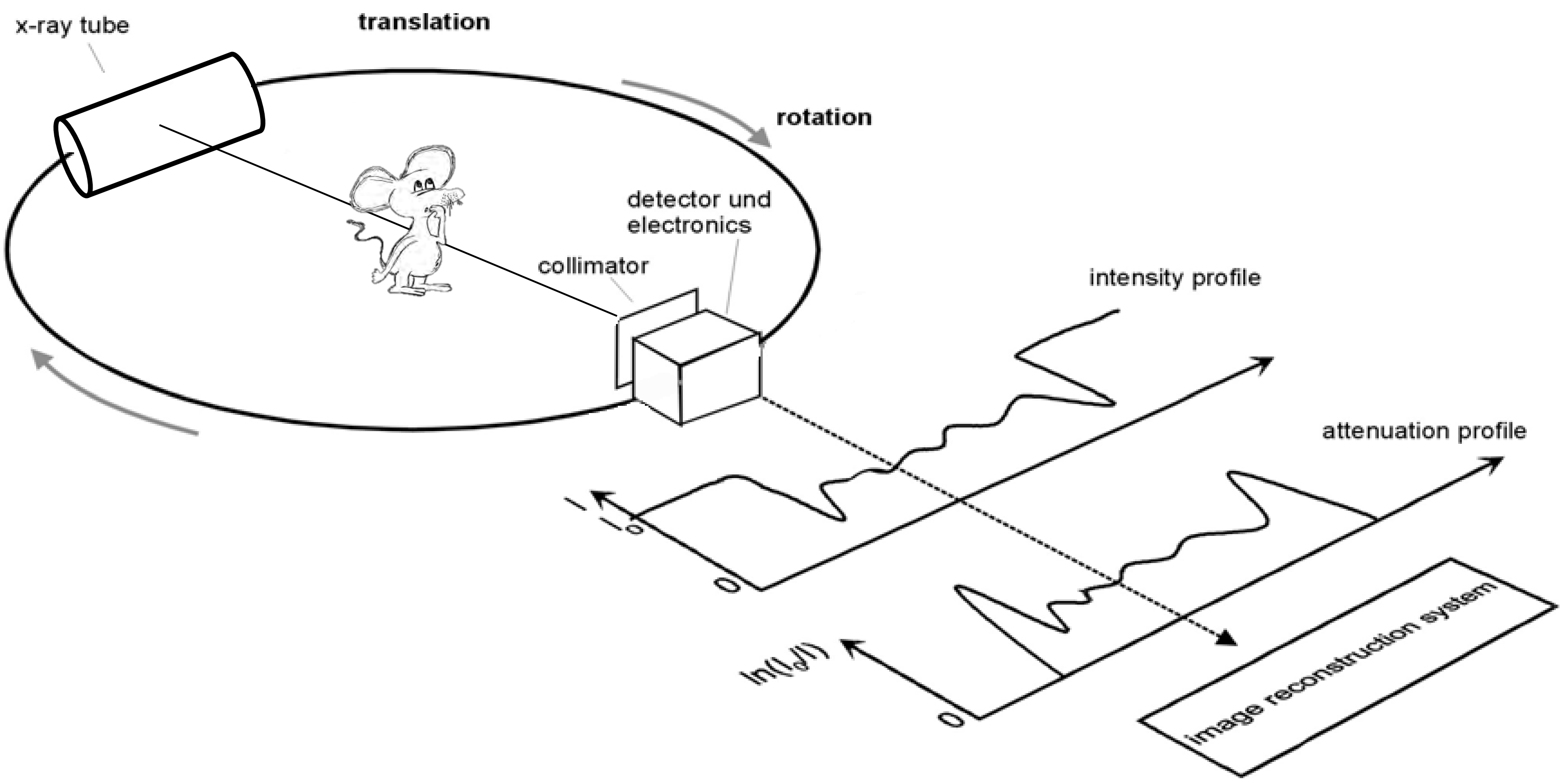
Computed X-ray tomography

CT Data Acquisition and Processing



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Computed X-ray tomography

microCT Reconstruction

No additional sorting process for projection data

→ already sinograms

Data will be corrected for detector inefficiencies and geometry distortions

→ system specific; similar to a PET normalization

microCT uses non-iterative methods similar to a FBP used in PET reconstruction

→ cone beam recon, Feldkamp

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ComputedX-ray tomography

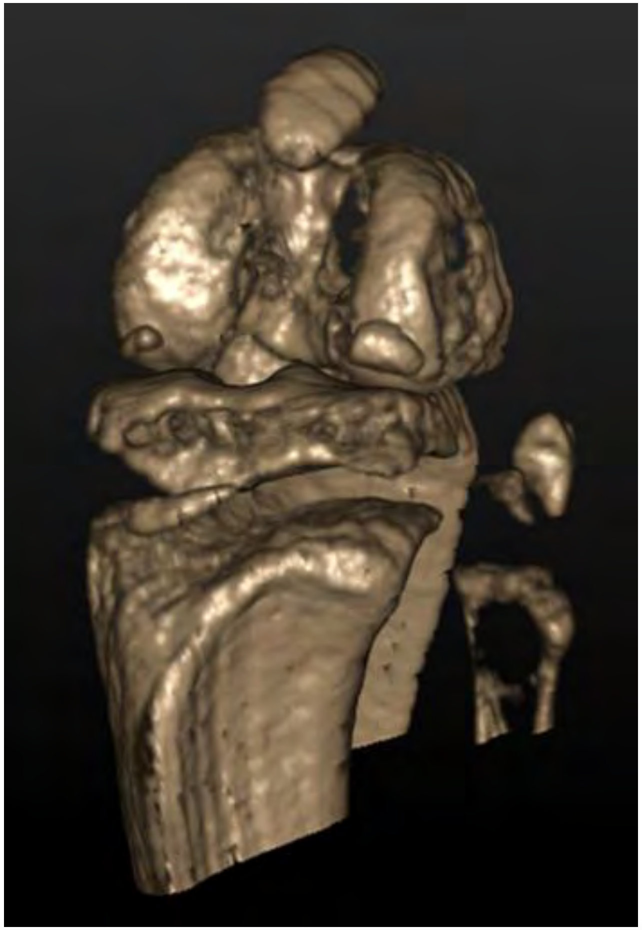
Bone Biology using CT

Easy to capture whole body CT

Digital zoom for high resolution CT imaging

Bone size and volume quantifiable via PMOD

High resolution renderings of mouse knee achievable through <35 µm voxels



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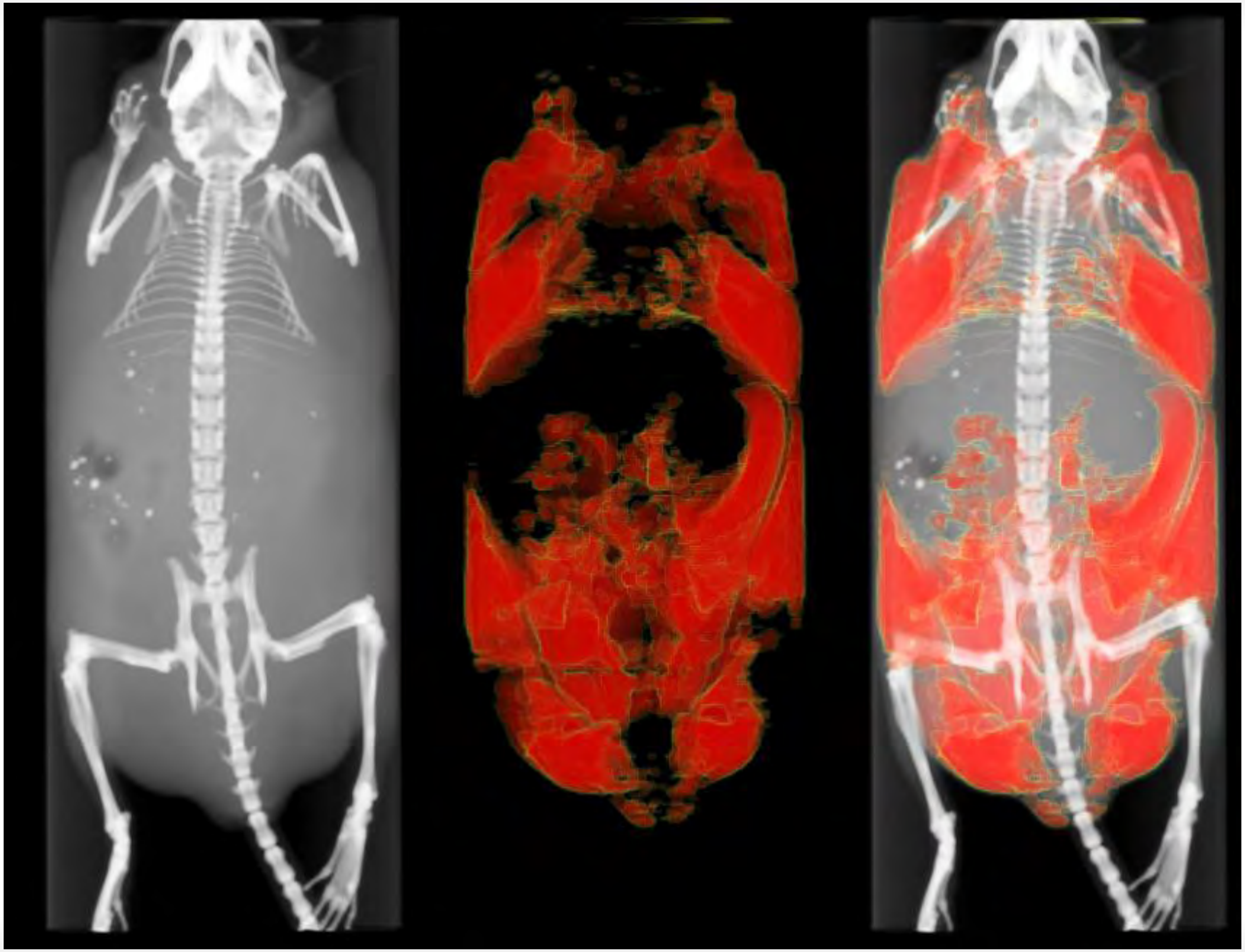
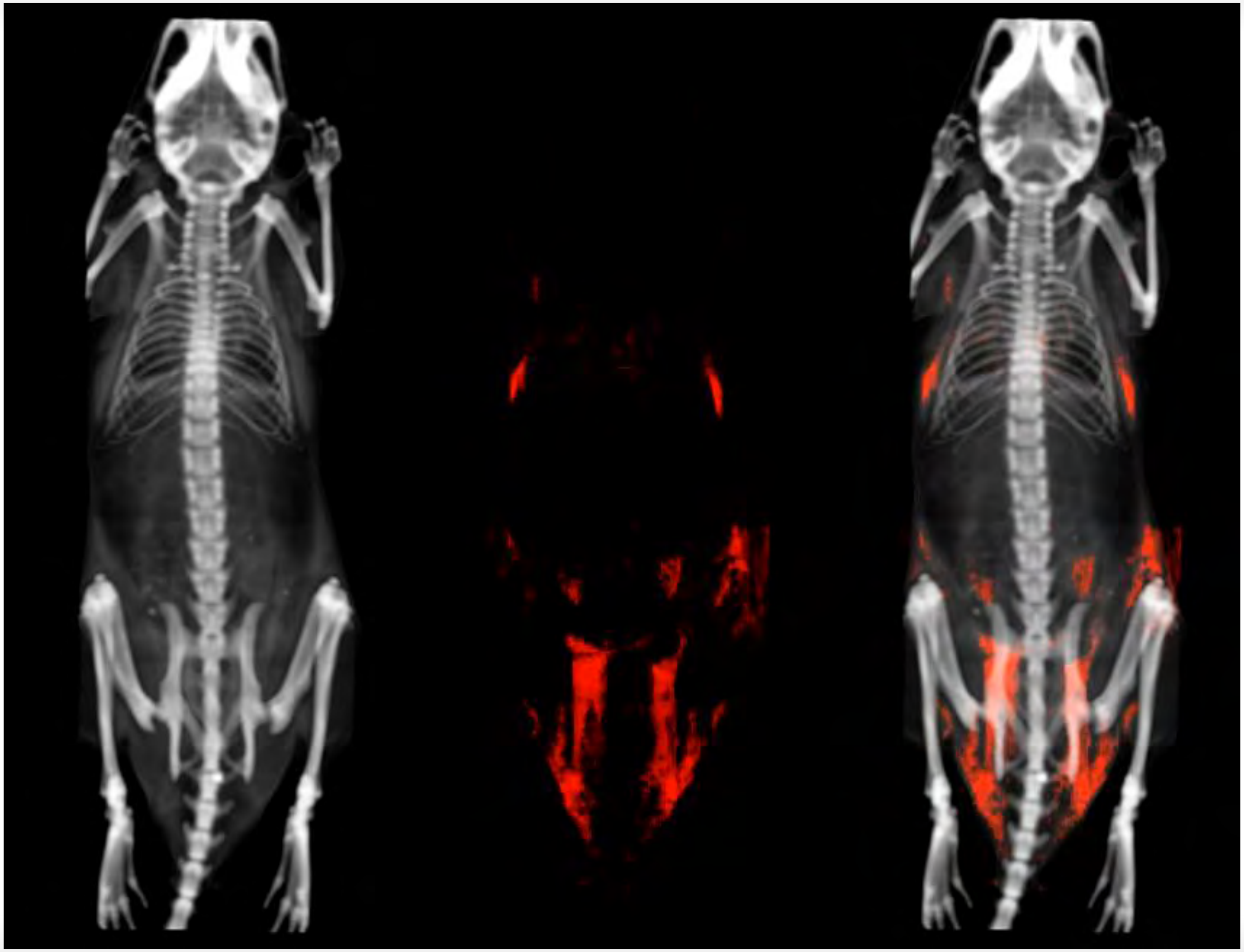
Computed X-ray tomography

Segmentation of adipose tissue – obese mouse



Computed X-ray tomography

Segmentation of adipose tissue – normal mouse



Computed X-ray tomography

Analysis of segmented fat areas

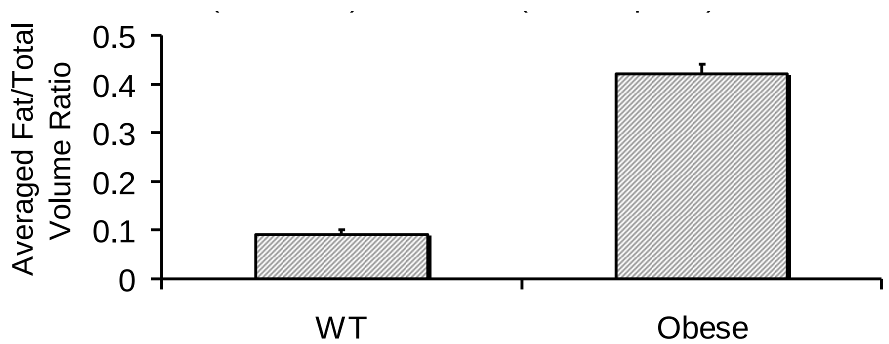
WT (C57BL/6J)

	Total (cm^3)	Fat (cm^3)	Fat / Total
Animal 1	28.79	3.00	0.10
Animal 2	33.25	3.05	0.09
Animal 3	30.30	2.63	0.09

Obese (B6.V-Lep^{ob}/J)

	Total (cm^3)	Fat (cm^3)	Fat / Total
Animal 1	66.25	26.75	0.40
Animal 2	61.15	26.31	0.43
Animal 3	64.19	25.7	0.40
Animal 4	54.25	23.78	0.44

Average Fat/Total volume ratios in
WT (C57BL/6J) and obese (B6.V-Lep^{ob}/J) mice



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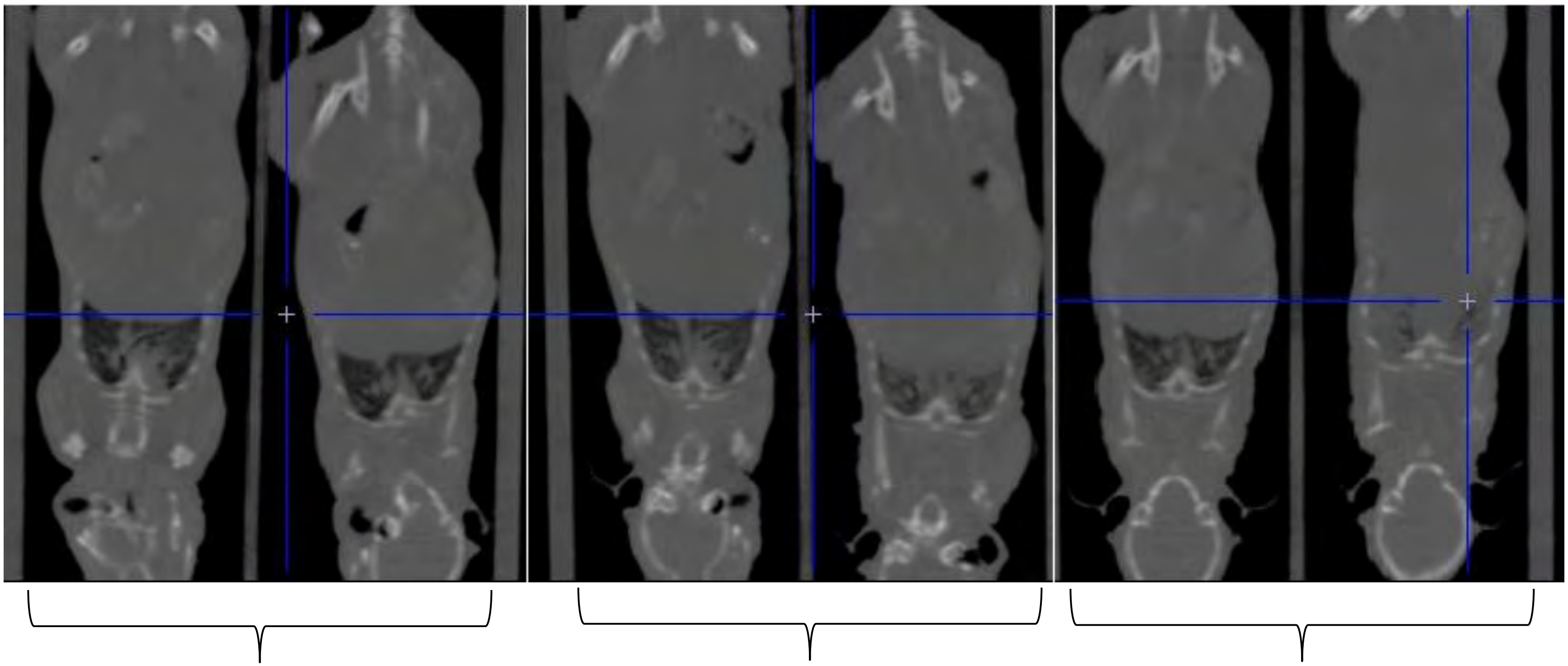
Computed X-ray tomography

Decrease in Lung Volume – Metastatic Mamacarcinom

M1 M2 M1 M2 M1 M2



Week 2 Week 4 Week 6

Animals imaged two at a time (Good resolution, 400 projections, 250 µm voxel size)

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Computed X-ray tomography

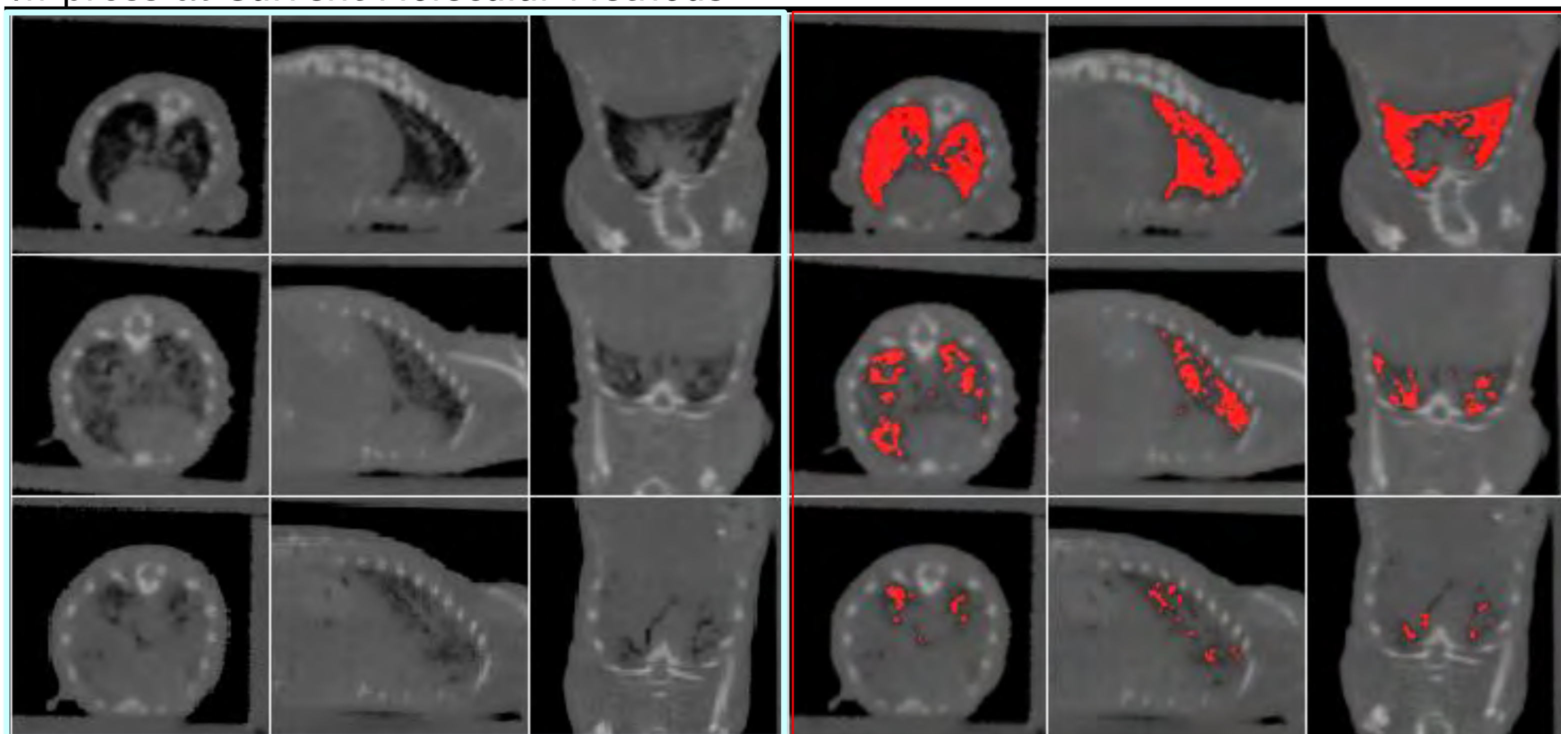
Tumor mediated Lung Degradation – Longitudinal Study

In press at *Current Molecular Methods*

WK 2

WK 4

WK 6



X-ray CT

Healthy Lung overlay on CT

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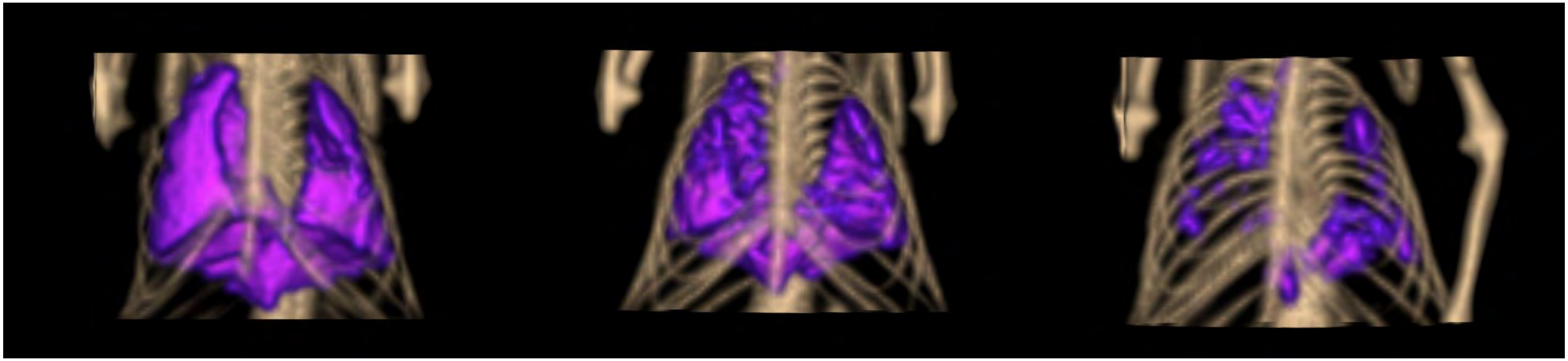
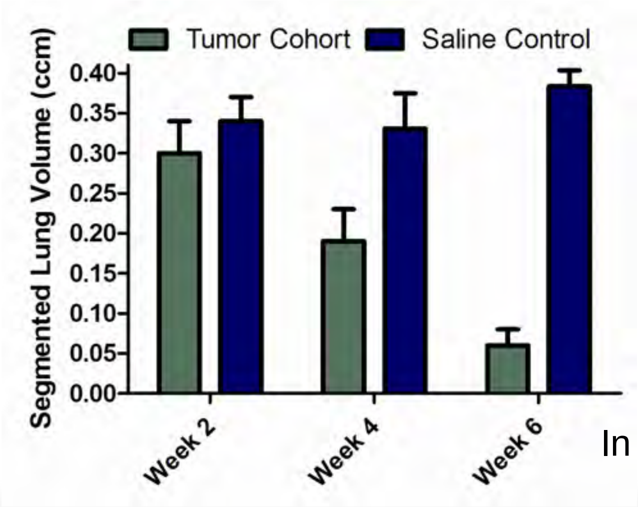
Computed X-ray tomography

Healthy Lung Degradation – Longitudinal Study

Week 2 Week 4 Week 6





In press at *Current Molecular Methods*

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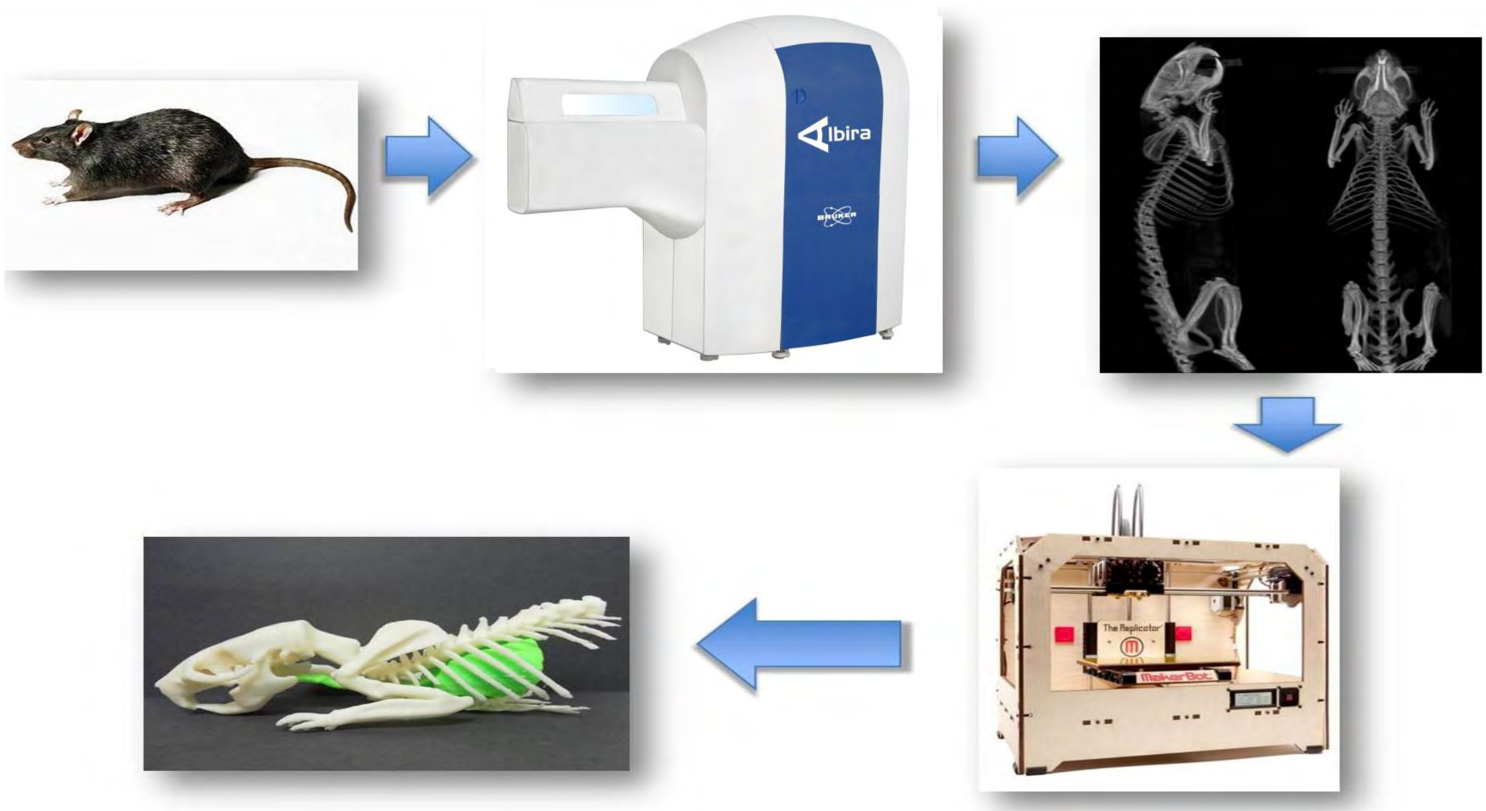
Computed X-ray tomography

3D Printing of preclinical X-ray CT Data Sets



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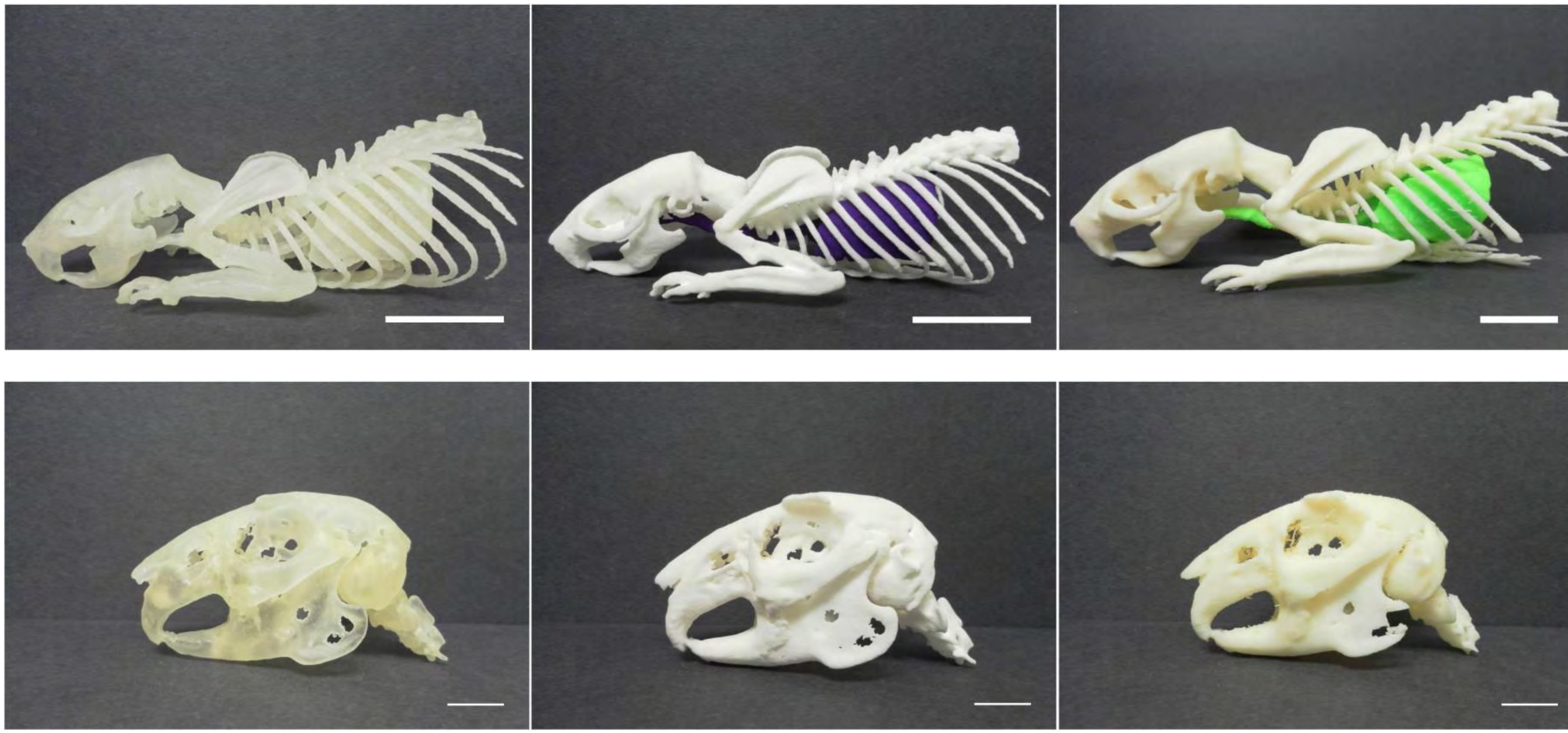
Computed X-ray tomography

3D Printing of preclinical X-ray CT Data Sets

Rat Skeleton and Lungs

Rabbit Skull



ProJet 3000 Overnight

Shapeways Inc.

MakerBot

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Computed X-ray tomography

CT Contrast Agents

Radiographic Contrast Agents:

Any substance that renders an organ or structure more visible than is possible without its addition.

Allows visulization of structures that can not be seen well or at all under normal circumstances.

Radiographic Contrast Agents are needed because:

soft tissue has a low absorption/interaction ratio

Absorption depends on:

- atomic number
- atomic density
- electron density
- part thickness
- K-shell binding energy (K-edge)

Computed X-ray tomography

CT Contrast Agents – Why does it work?

	Atomic Number	Atomic Density	Electrons / cm^3	Main Element
Air	7.64	0.00129	0.0039×10^{23}	Oxygen
Fat	5.92	0.91	3.27×10^{23}	
Water	7.42	1.00	3.34×10^{23}	Oxygen
Bone	13.8	1.85	5.5×10^{23}	Calcium
Iodine	53	4.93		Iodine
Barium	56	3.5		Barium

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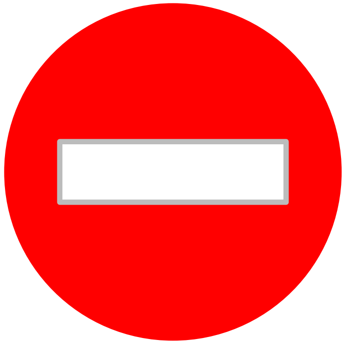
CT Contrast Agents – Types of Contrast Media

NEGATIVE

- Air
- Oxygen
- Carbon Dioxide
- Nitrous Oxide



POSITIVE

- Barium
- Iodine

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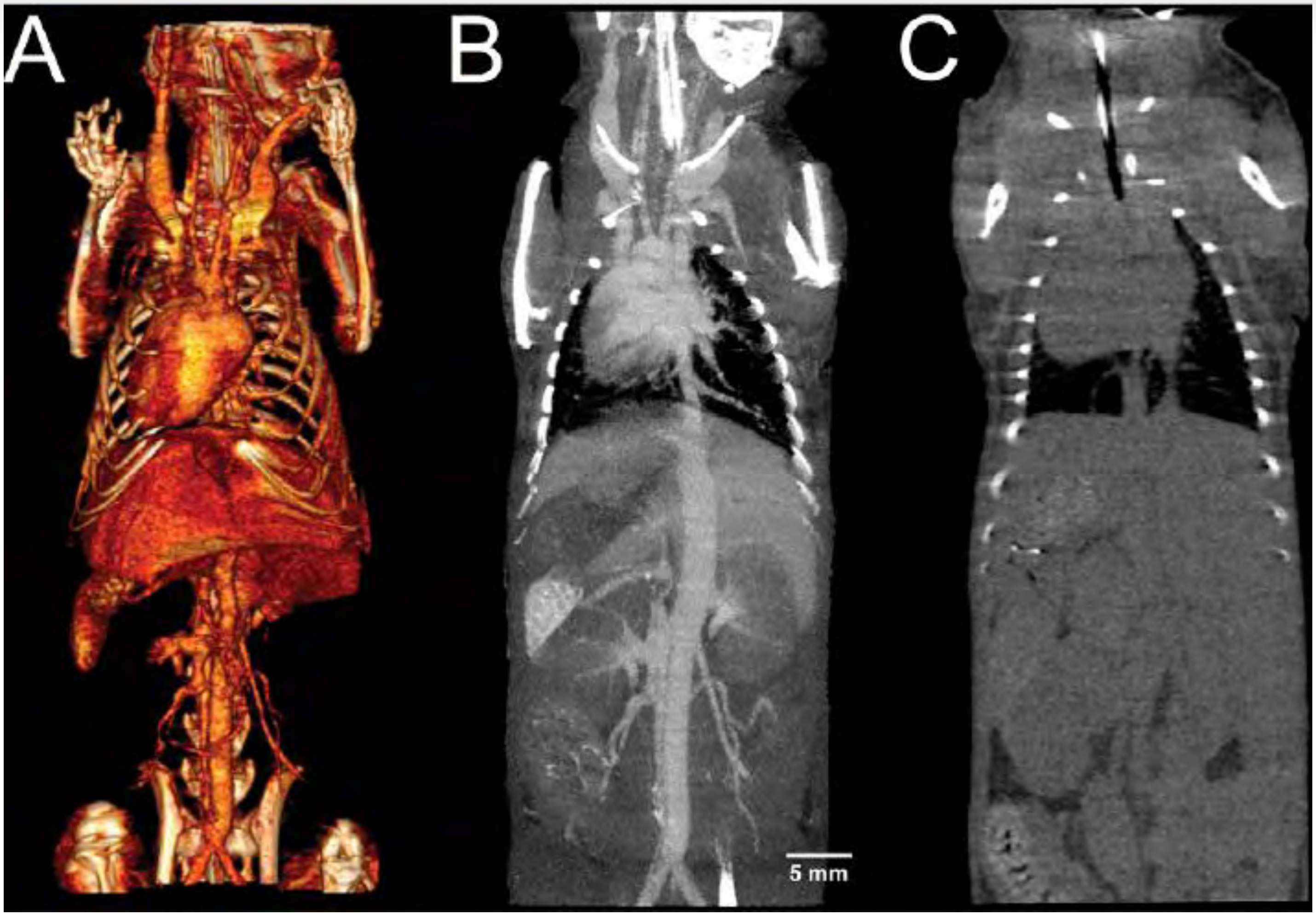
Computed X-ray tomography

Contrast Agent enhanced picture – Angiography, Liver & Spleen



Boll H, Nittka S, Doyon F, Neumaier M, Marx A, et al. (2011) Micro-CT Based Experimental Liver Imaging Using a Nanoparticulate Contrast Agent: A Longitudinal Study in MicePLoS ONE 6(9)

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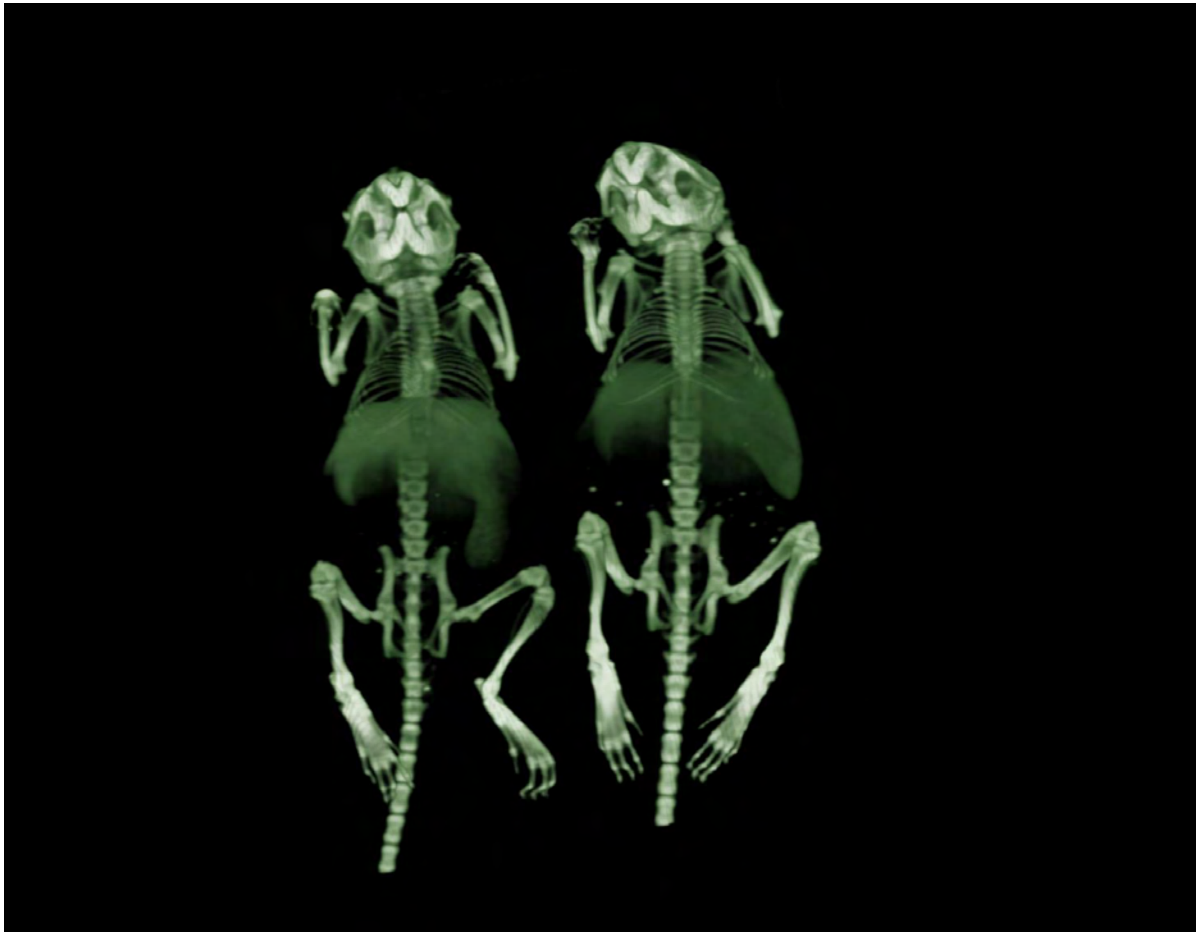
Computed X-ray tomography

Novel Liver Contrast Agent for CT



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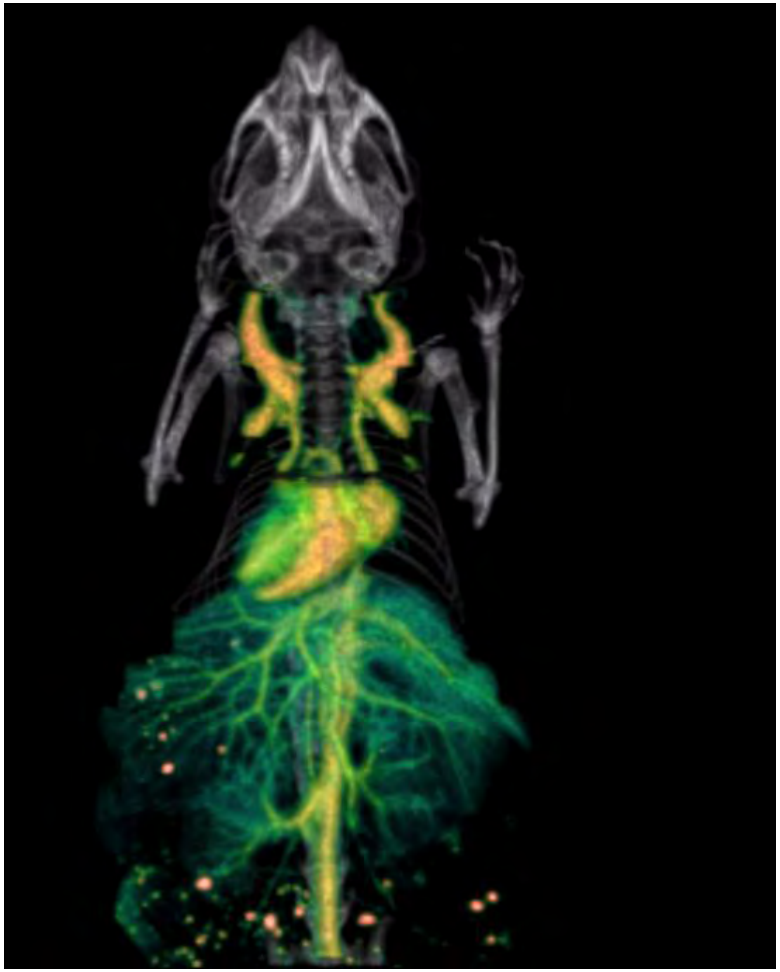
Computed X-ray tomography

Novel Contrast Agent for CT of Vascular System



Contrast Media = Aurovist

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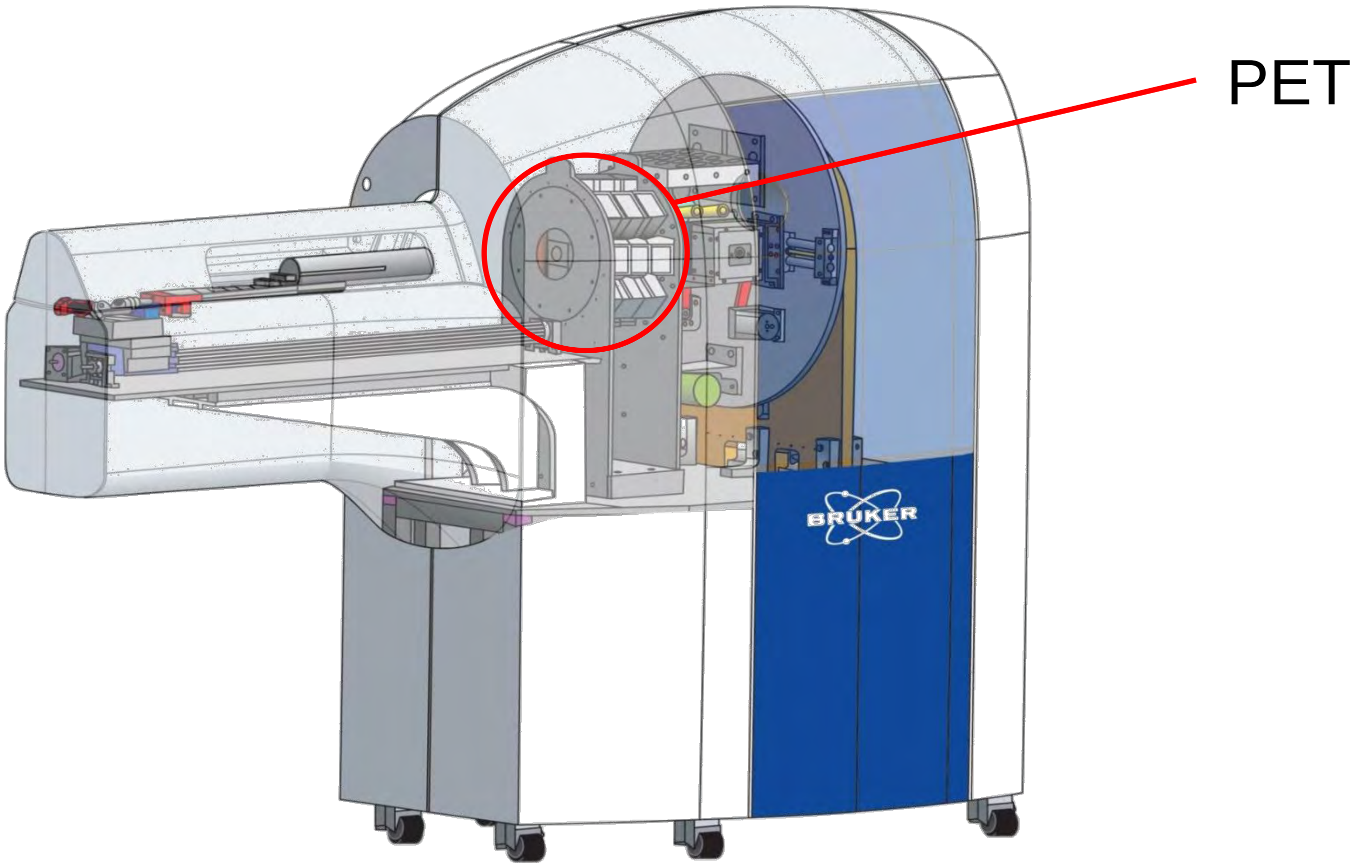
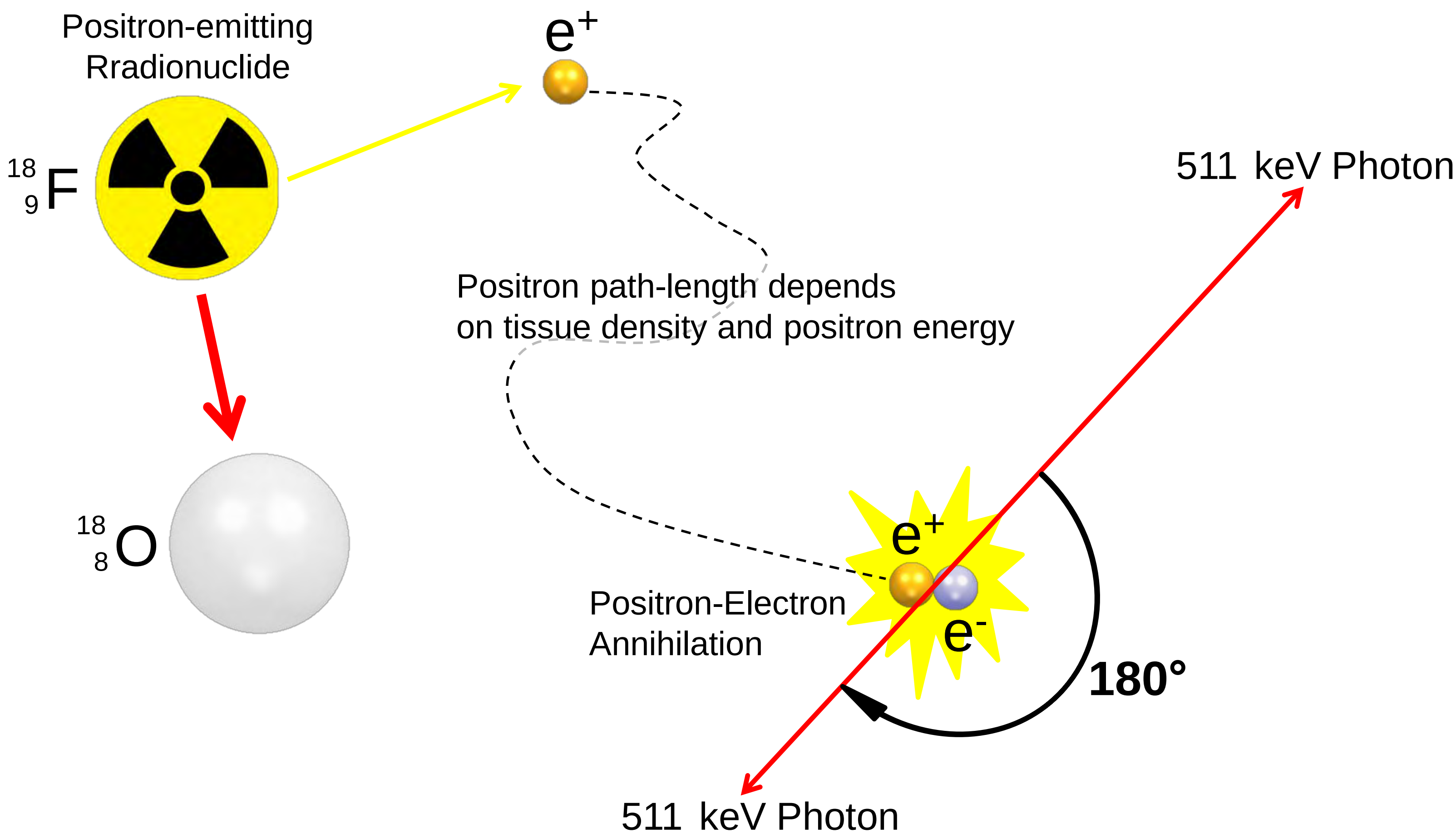
Positron Emission Tomography

PET



Positron Emission Tomography

Positron – Electron Annihilation



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Positron Emission Tomography

Positron Emitting Radionuclides

Isotope	Halflife	β^+ fraction	Max. Energy	range(mm)
C–11	20.4 mins	0.99	0.96 MeV	0.4 mm
N–13	9.96 mins	1.00	1.20 MeV	0.7 mm
O–15	123 secs	1.00	1.74 MeV	1.1 mm
F–18	110 mins	0.97	0.63 MeV	0.3 mm
Cu–62	9.74 mins	0.98	2.93 MeV	2.7 mm
Cu-64	12.7 hours	0.19	0.65 MeV	0.3 mm
Ga–68	68.3 mins	0.88	1.83 MeV	1.2 mm
Br-76	16.1 hours	1.00	1.90 MeV	1.2 mm
Rb–82	78 secs	0.96	3.15 MeV	2.8 mm
I–124	4.18 days	0.22	1.50 MeV	0.9 mm

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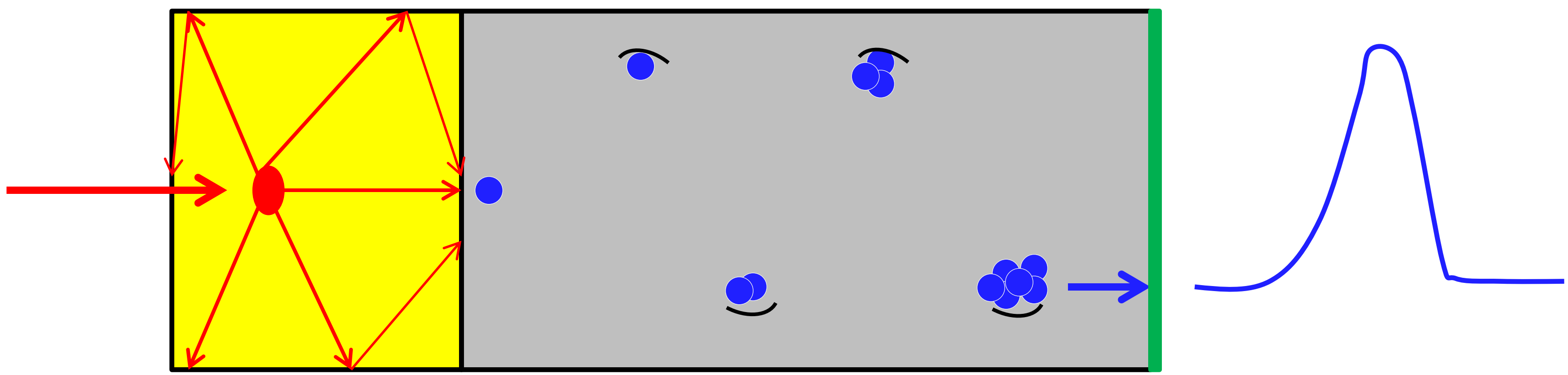
Positron Emission Tomography

ALBIRA Gamma-ray Detector Principle

PSPMT
position sensitive photomultiplier tube



Single
continuous crystal

Advanced Detector
Electronics

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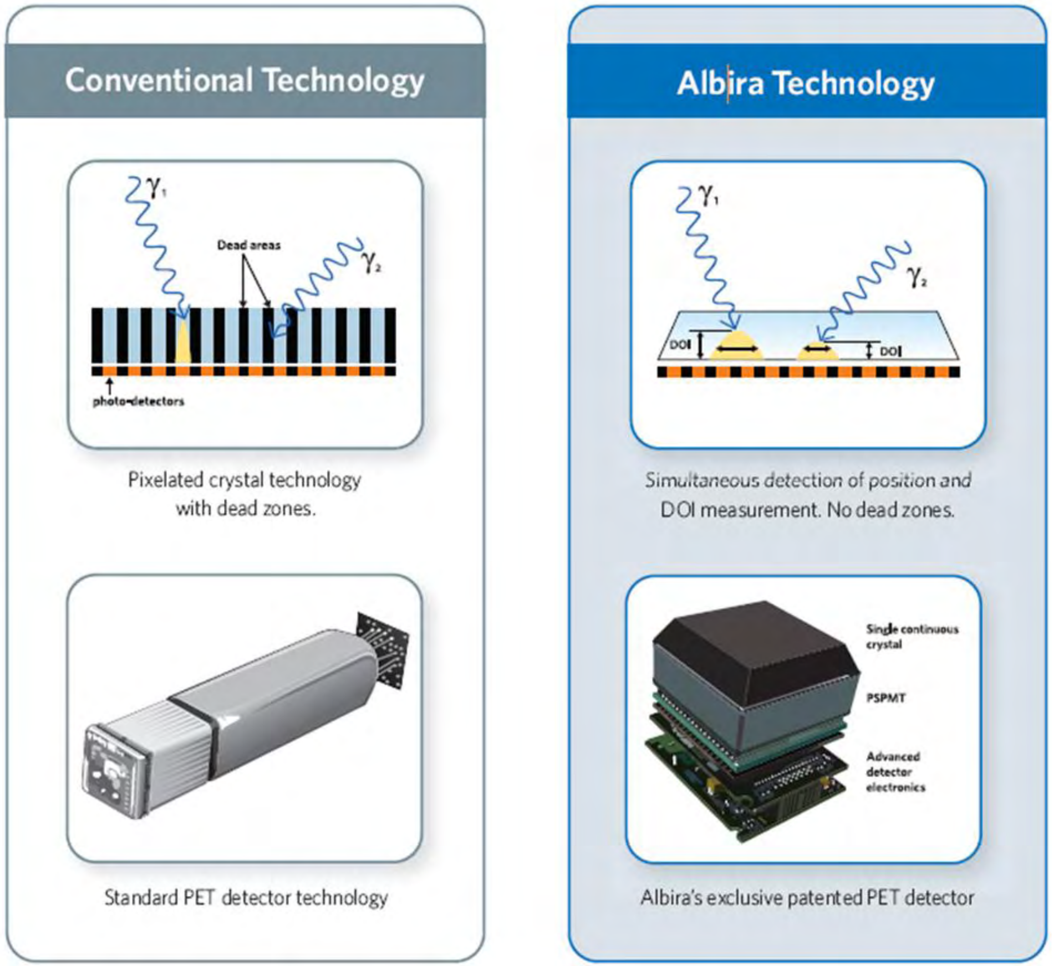
Positron Emission Tomography

Positron – Electron Annihilation



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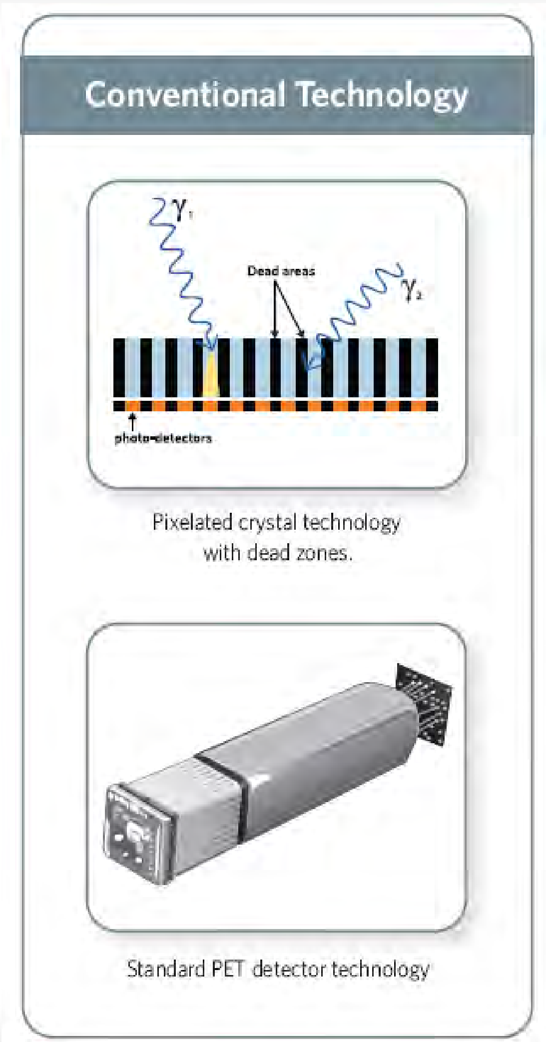
Positron Emission Tomography

Positron – Electron Annihilation



Current technology utilized packed crystals with dead zones

Tighter packing yields more dead zones

Susceptible to the parallax error

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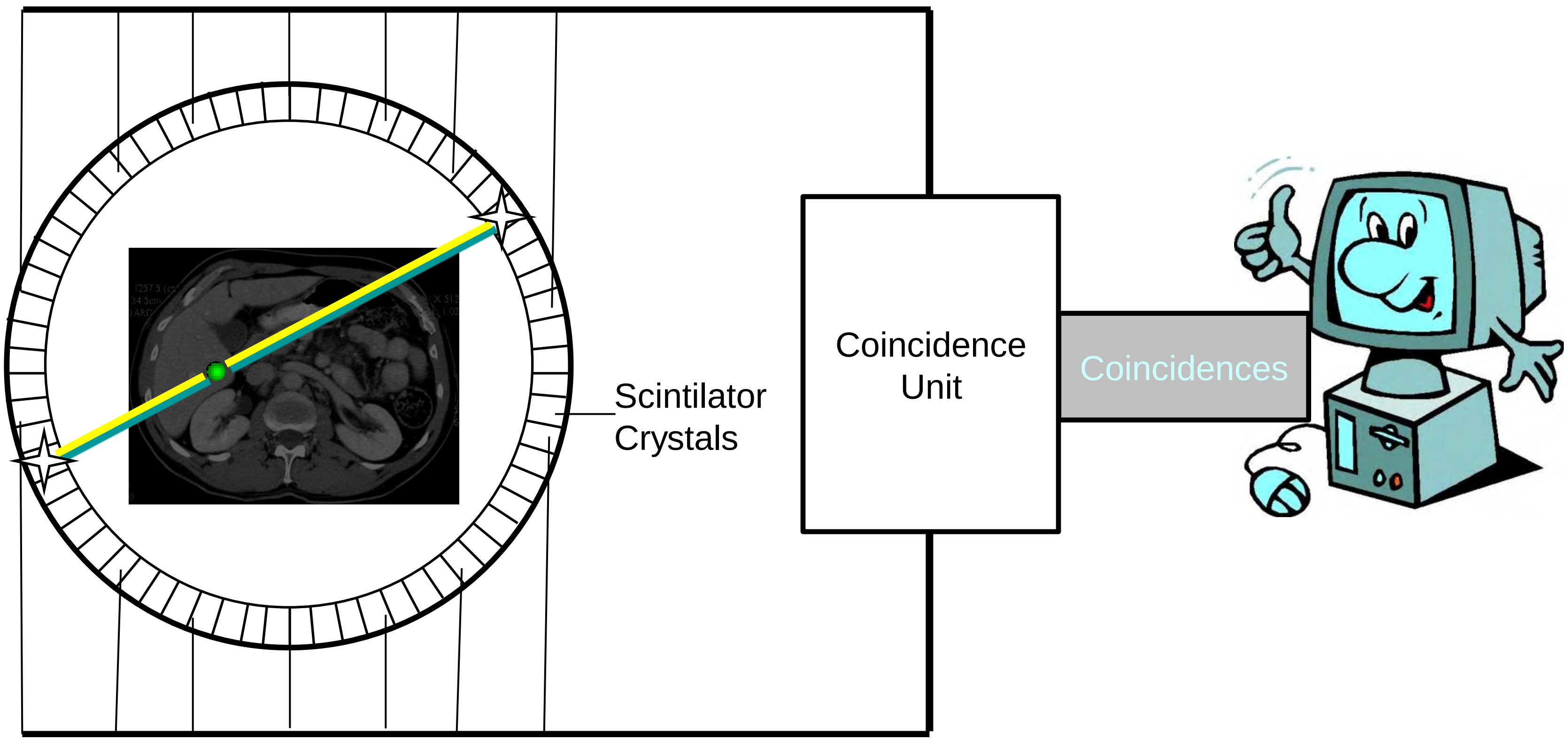
Positron Emission Tomography

Operation of a PET-Scanner



Positron Emission Tomography

ɣ-ray Detection in a PET system

True Coincidences

both ɣ-rays escape without scatter and interact in detctors

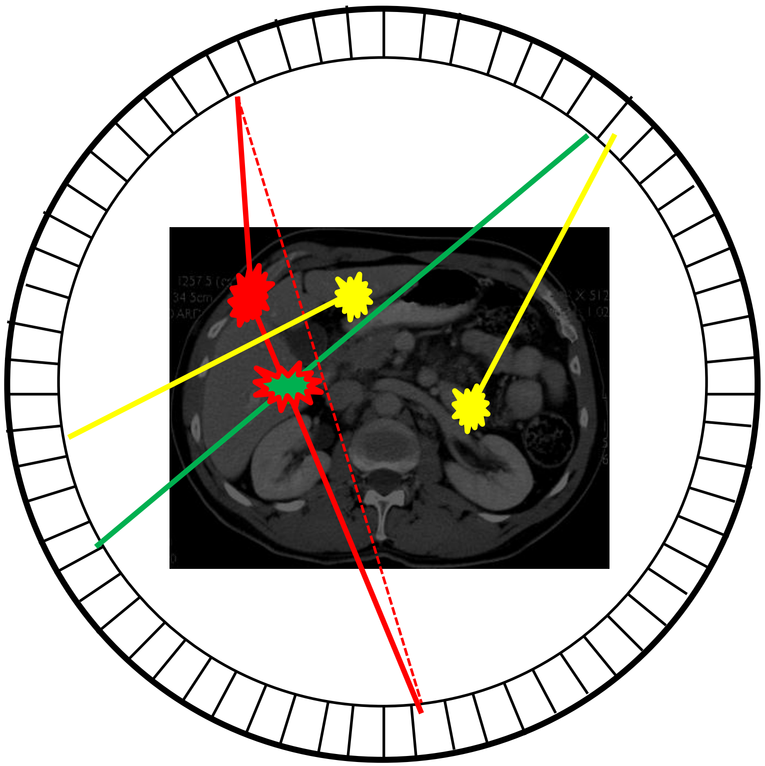
Scatter coincidences

one, or both ɣ-rays scatter in tissue

Random coincidences

two ɣ-rays from different origins strike the detectors at the same time

(a.k.a. *accidental coincidences*)



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Positron Emission Tomography

PET Hardware

Scintilators

- High stopping power
- High light output
- Fast scintillator
- Small crystal size

→ High spatial resolution

LSO, LYSO, YAP, etc.

Light-Detectors

Photomultiplier Tubes (PMT)

- Single Channel
- Multi Channel

Solid State Detectors

- Avalanche Photo Diodes (APD)
- Geiger-Mode APDs Silicon-PMTs

Detectortype

- Single Crystal Coupling
- Block Detector
- Detectors with DOI capabilities (Phoswitch)

- A full PET system comprises several detector rings summing up to several 1000 to 10.000 individual crystals
- The performance of a PET system as well as physical limitations will be determined by the choice of hardware

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Positron Emission Tomography

Important Scanner Parameters

Energy Resolution

detection limit for measured energy of detected ɣ-rays

Timing Resolution

time variation (inaccuracy) of the system for detection of two single events originating from the same annihilation

Spatial Resolution

smallest object that can be visualized (partial volume effect)

Sensitivity

detection limit for radiotracer (isotope) or contrast media

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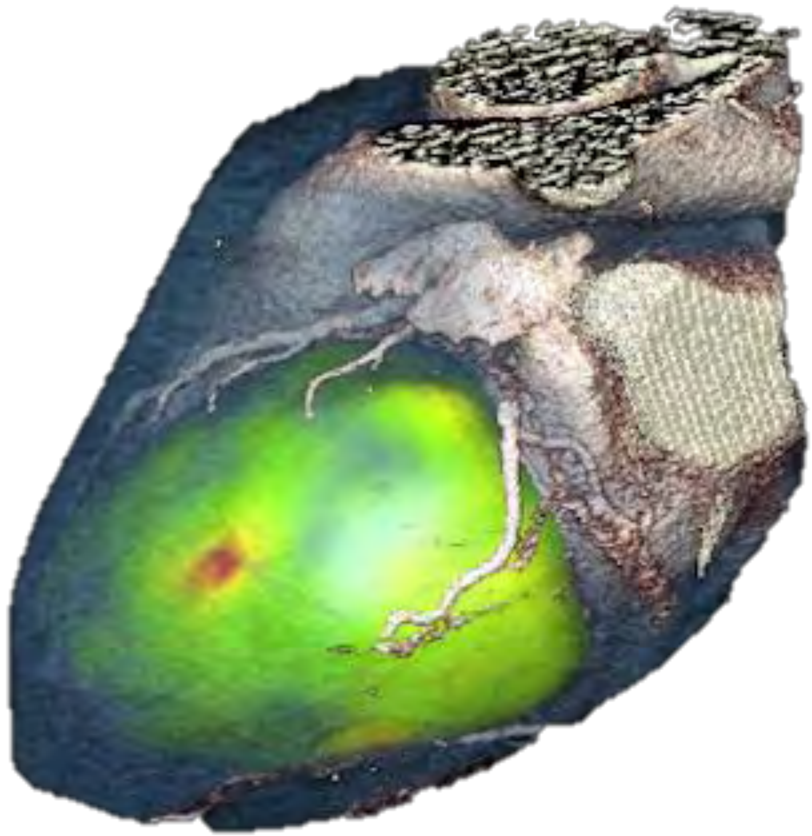
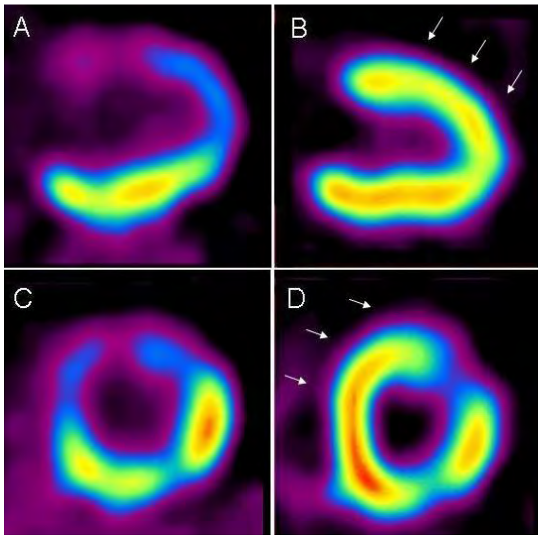
Positron Emission Tomography

Cardiology

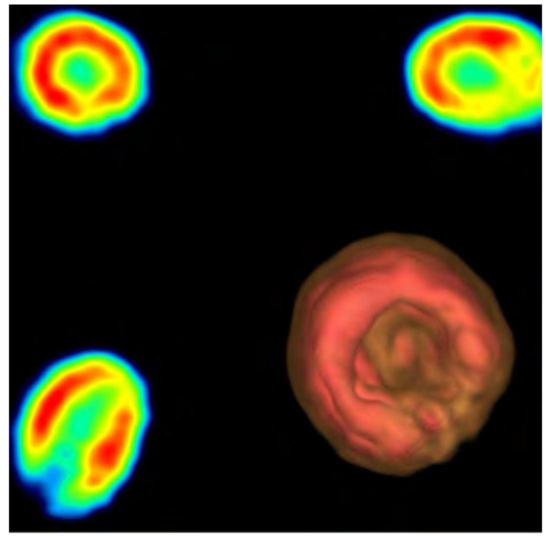


Positron Emission Tomography

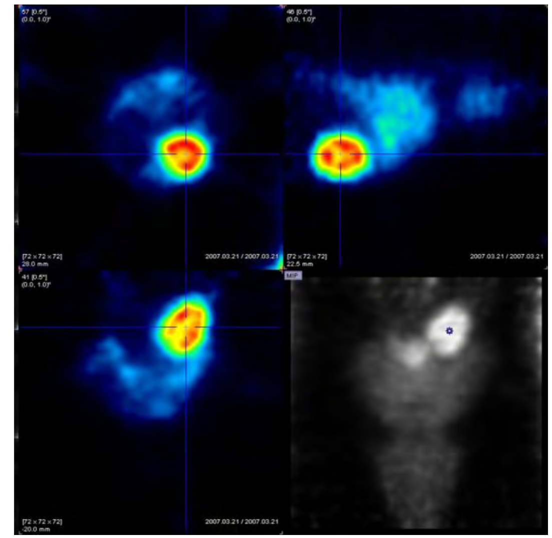
Cardiology – From mouse to man



^{18}F-FDG – human heart [A]



^{18}F-FDG –rat heart [B]



^{18}F-FDG – mouse heart

[A] Tossios, P., et al. No evidence of myocardial restoration following transplantation of mononuclear bone marrow cells in coronary bypass grafting surgery patients based upon cardiac SPECT and 18F-PET. BMC Medical Imaging. (2006),

[B] Courtesy of Prof. J. Viña, Research Unit, Physiology Faculty, Uni. Valencia.

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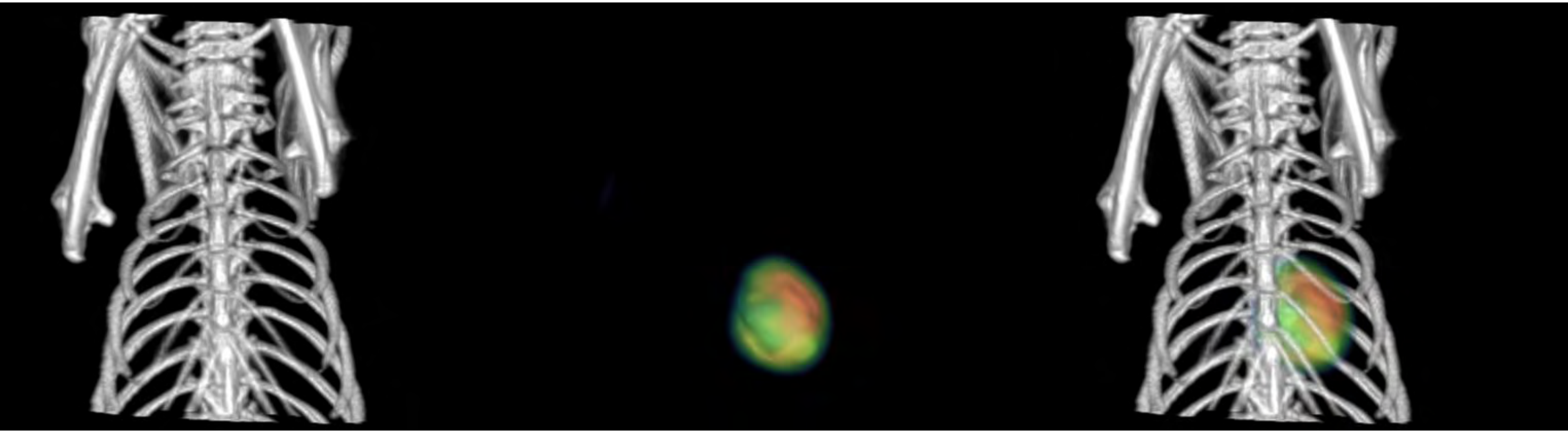
Positron Emission Tomography

Cardiology - 3D imaging of a rat heart (PET/CT-Fusion)



^{18}F-FDG imaging of a rat heart without gating

Major walls of the heart are clearly visible

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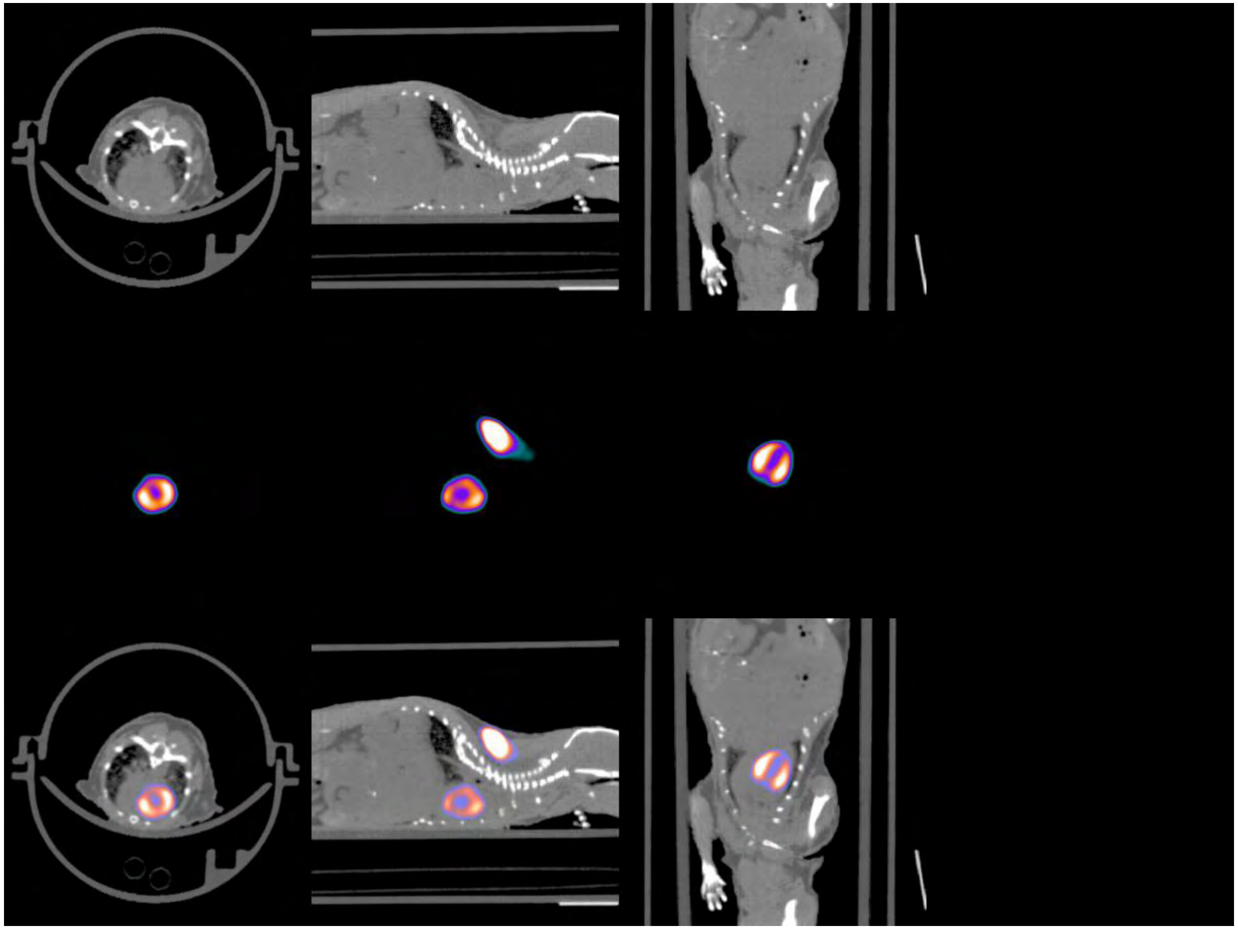
Positron Emission Tomography

Cardiology - Mouse heart imaging - without gating



Positron Emission Tomography

Cardiology - Mouse heart imaging - without gating

- With a high resolution PET scanner gating may not always be necessary
- Saves time and dose
- Protocol
  - 500 uCi of 18F-FDG injected
  - Data acquired for 10 min
  - Data reconstructed with MLEM
  - Data fused with CT

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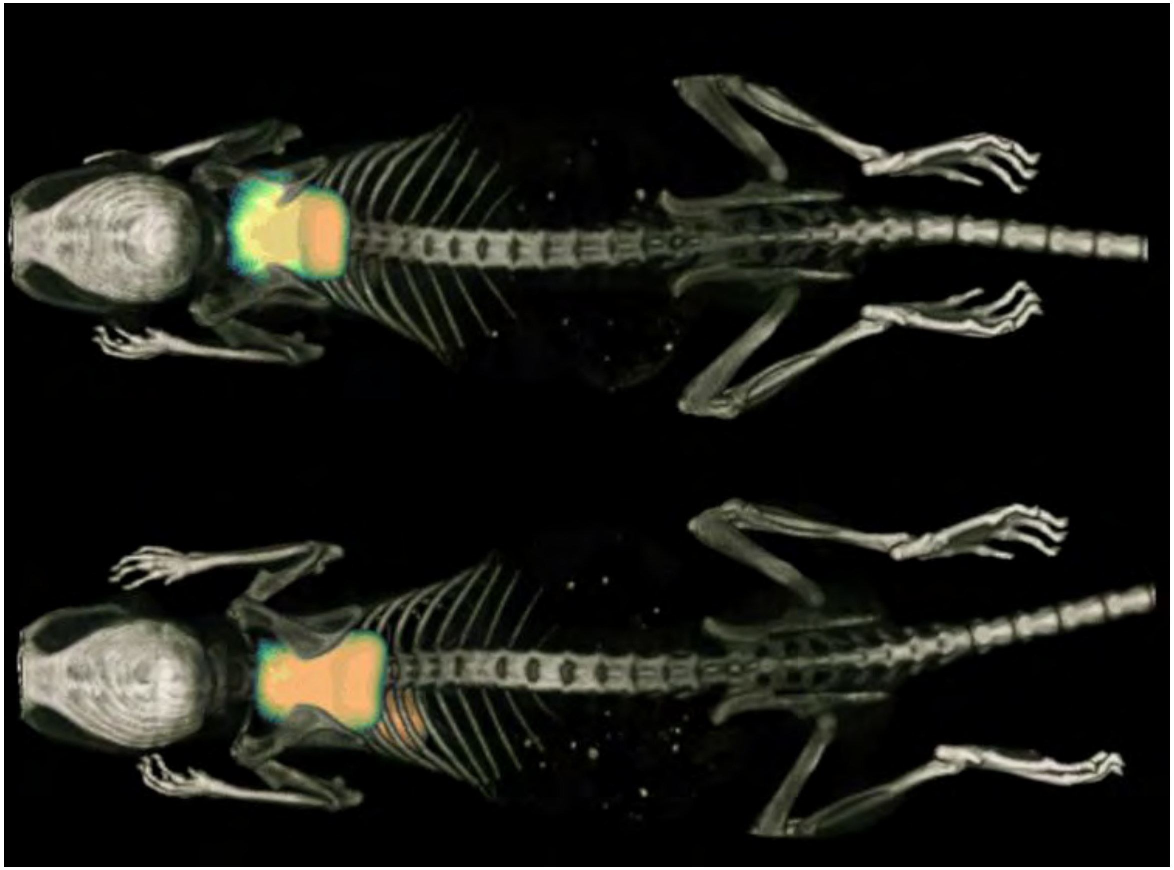
Positron Emission Tomography

Metabolic Diseases



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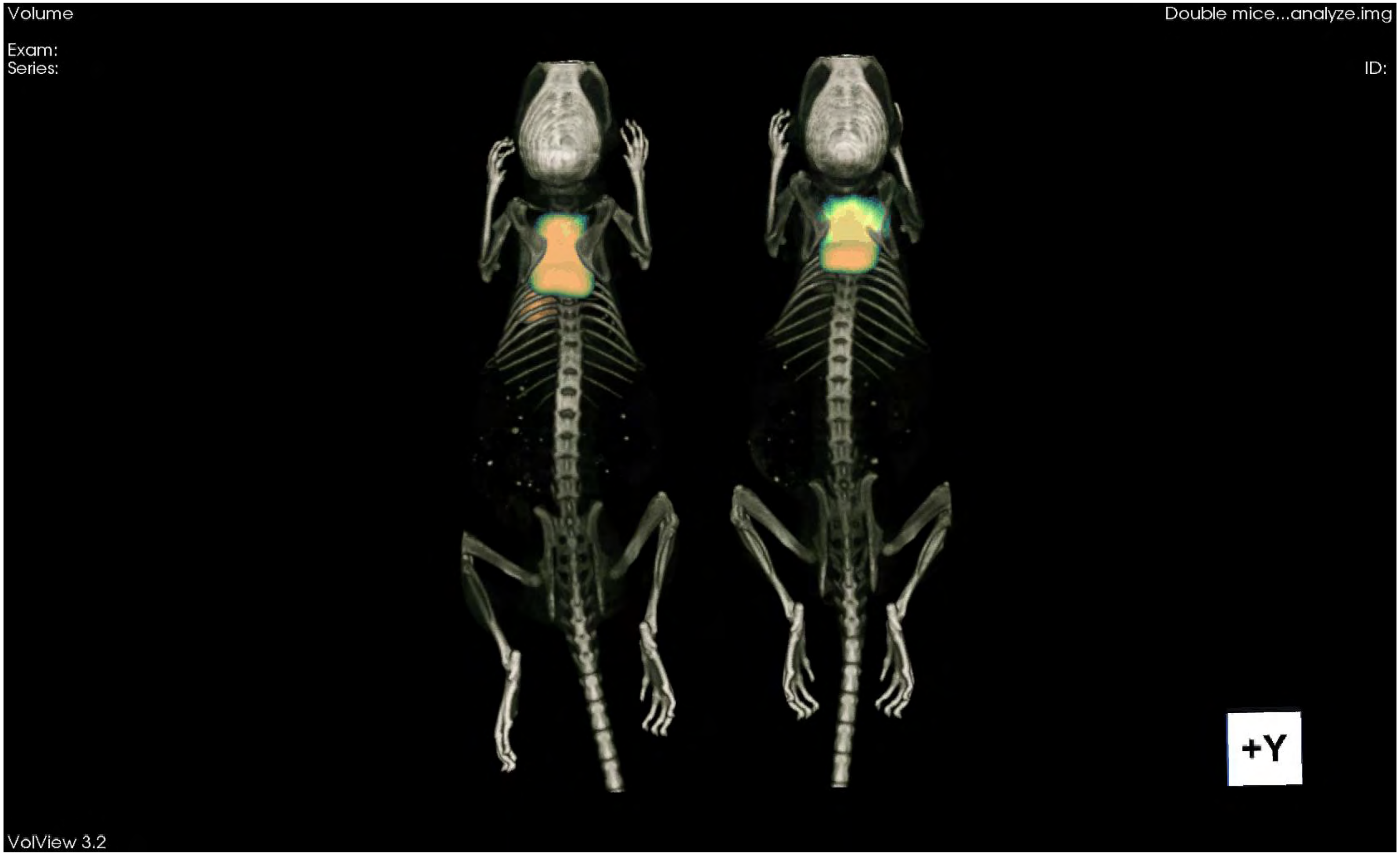
Positron Emission Tomography

Brown fat in mice – PET/CT-fusion



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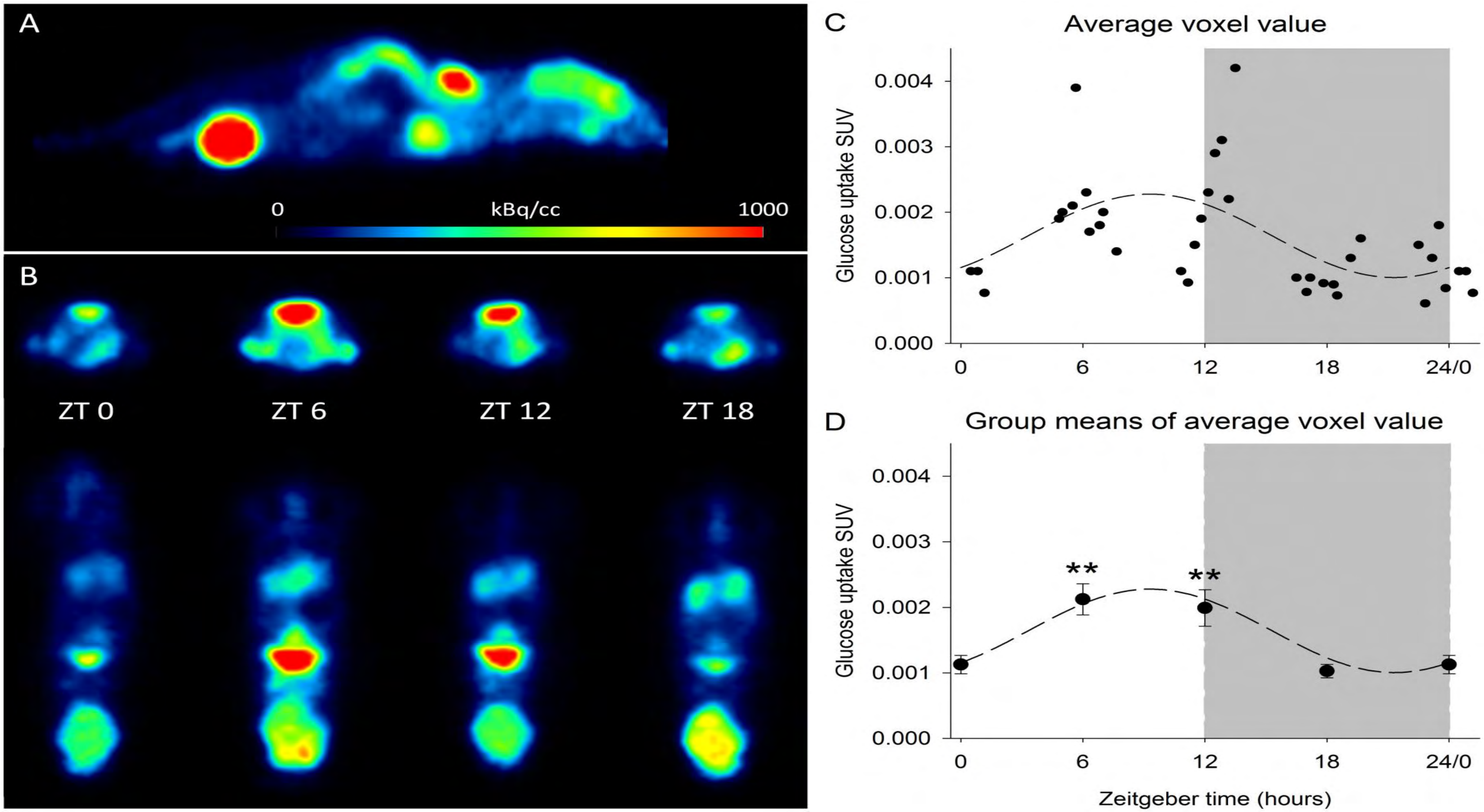
Positron Emission Tomography

Brown fat in mice – PET/CT-fusion



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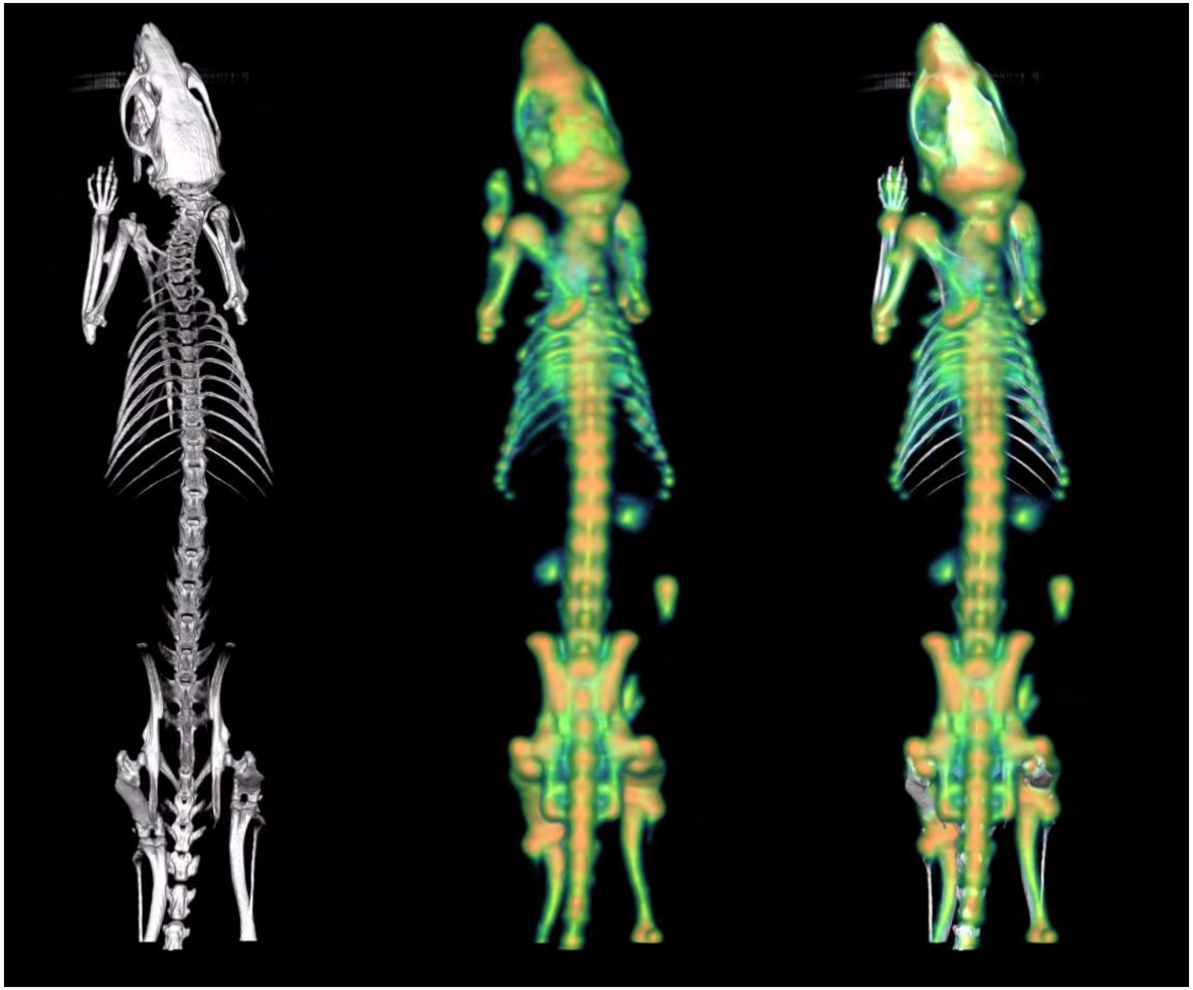
Positron Emission Tomography

Bone Development and Bone Diseases

Positron Emission Tomography

^{18}F-NaF PET/CT imaging in rat



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Positron Emission Tomography

Bone Development in mouse – single PET scan

- Injected 200 uCi of ^{18}F NaF
- Imaged on Albira 2 ring PET/CT
- Areas of new bone growth show significant uptake

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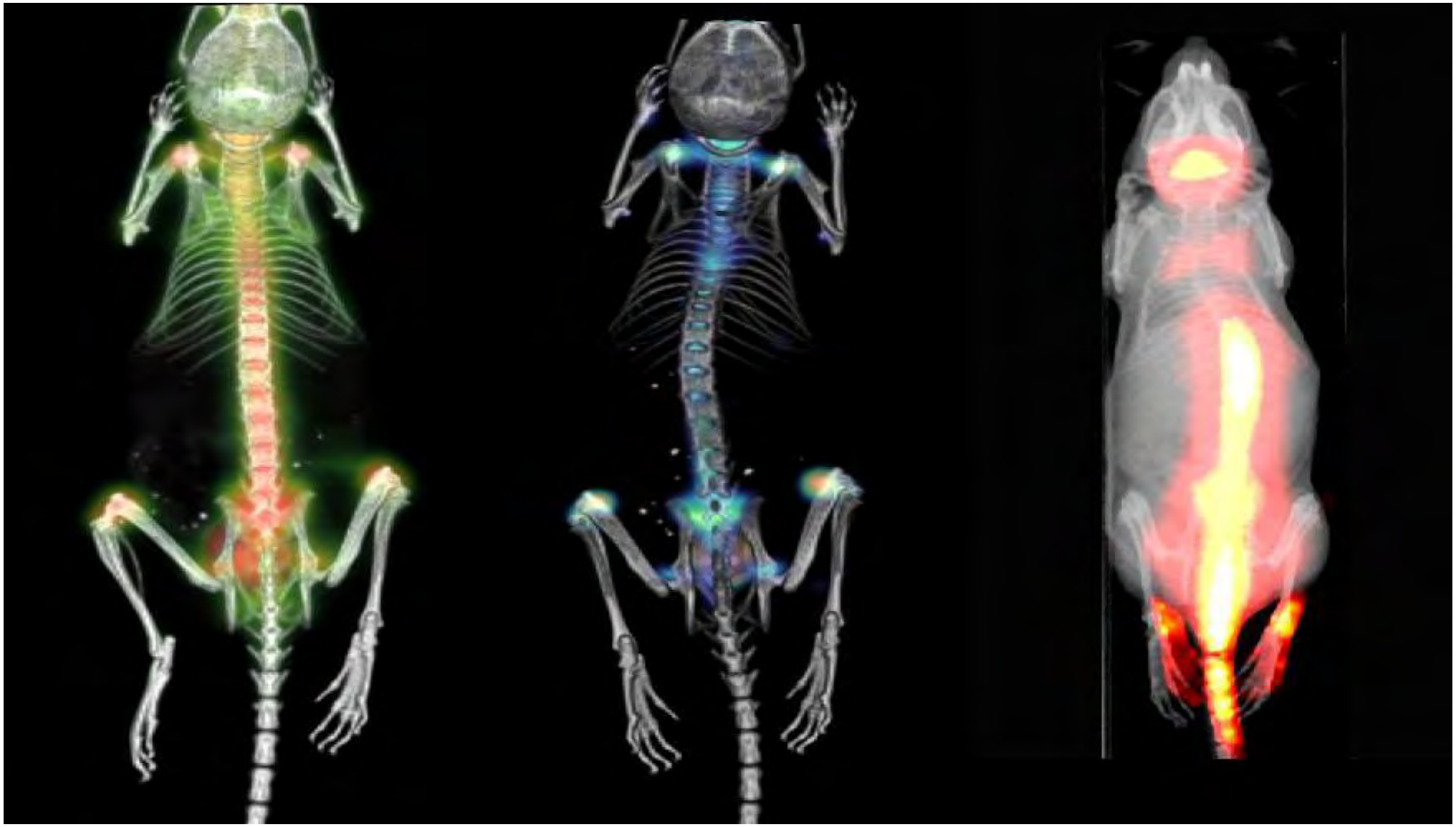
Positron Emission Tomography

Triple Tracer Imaging – PET/SPECT/Optical



PET/CT
($Na^{18}F$)

SPECT/CT
(^{99m}Tc-MDP)

Fluorescence/X-ray
(Osteosense-750)

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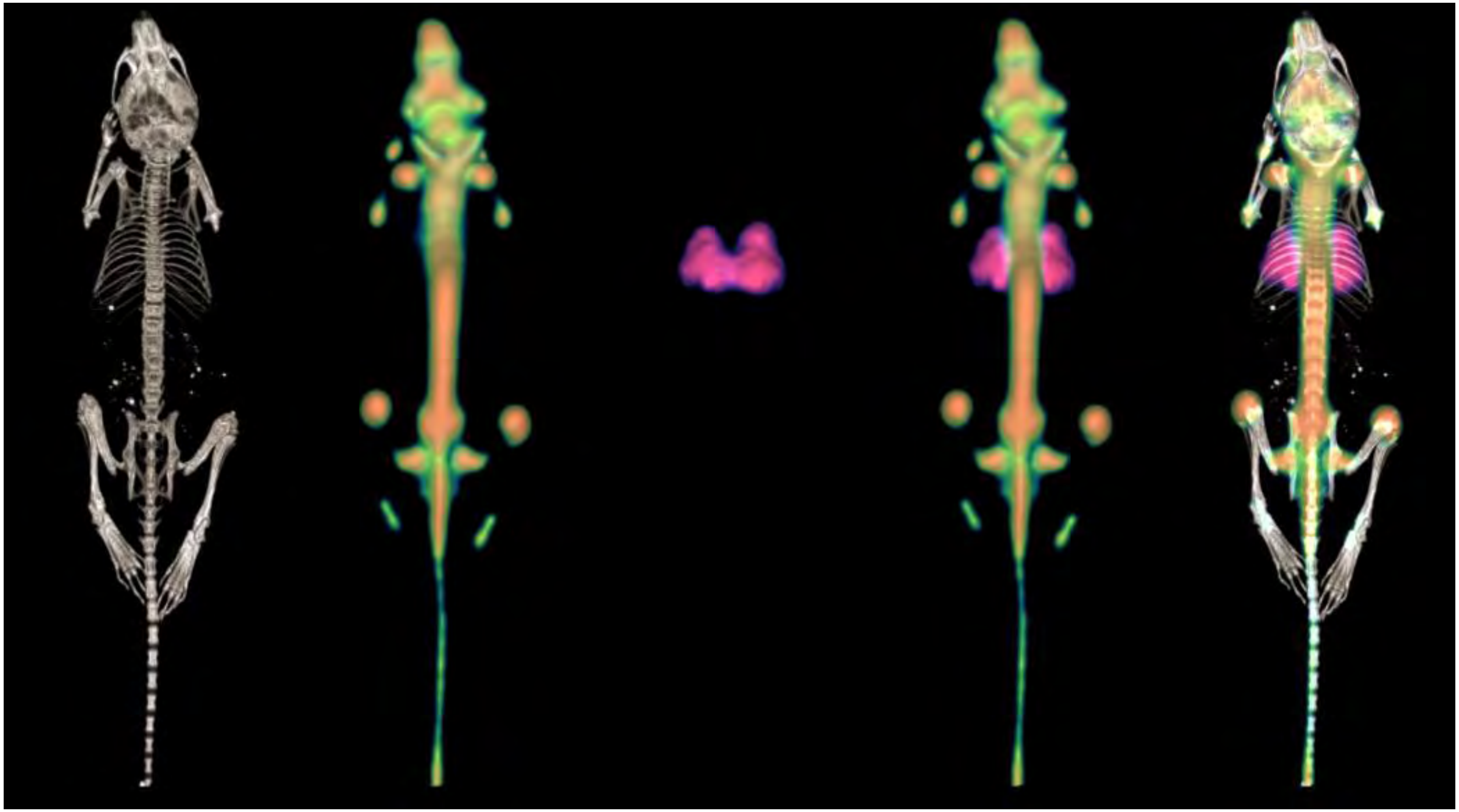
Positron Emission Tomography

Dual Tracer Mouse Scan – PET/SPECT/CT



CT

PET ($Na^{18}F$)

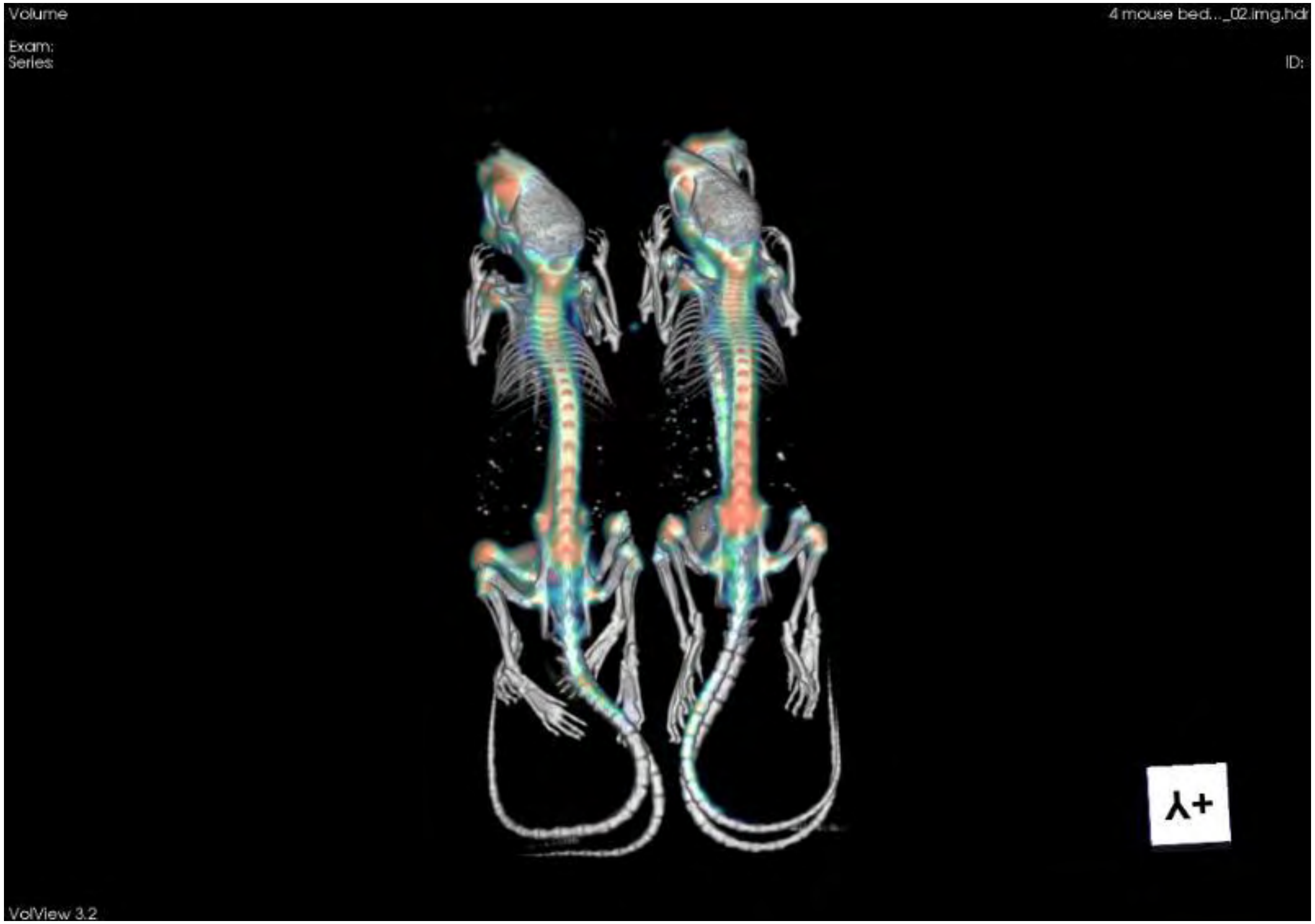
SPECT (^{99m}Tc-MAA)

PET/SPECT Overlay

Trimodal Overlay

Positron Emission Tomography

^{18}F-NaF – Four Mice PET/CT



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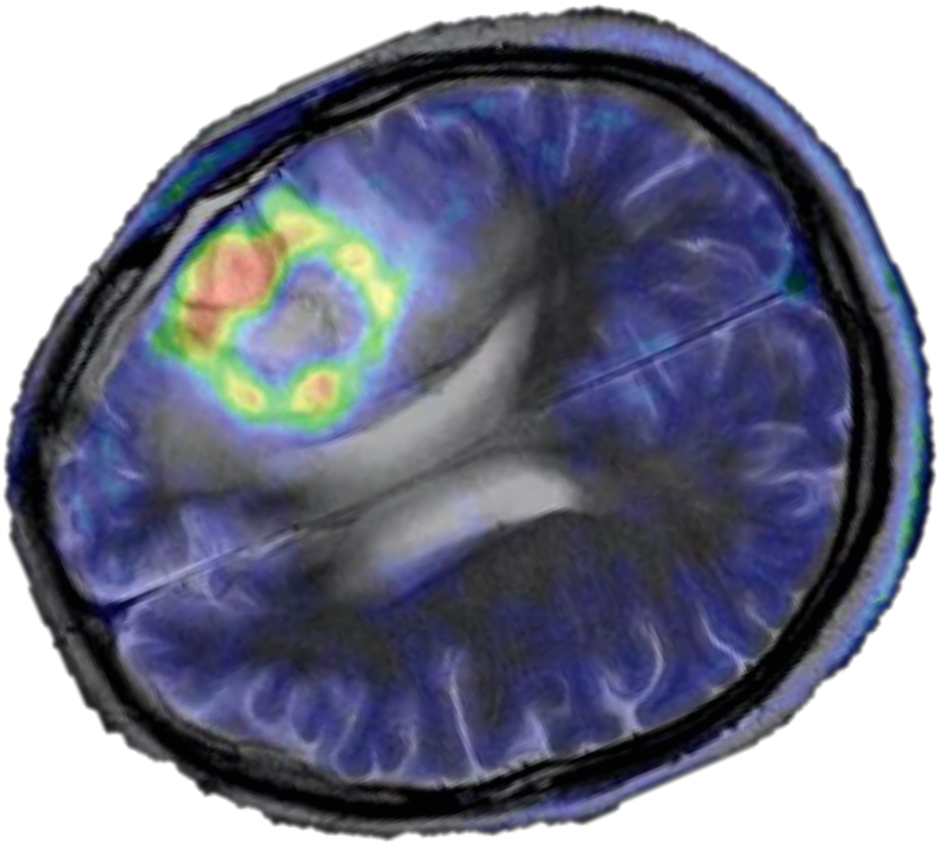
Positron Emission Tomography

Neurology – Functional Brain imaging



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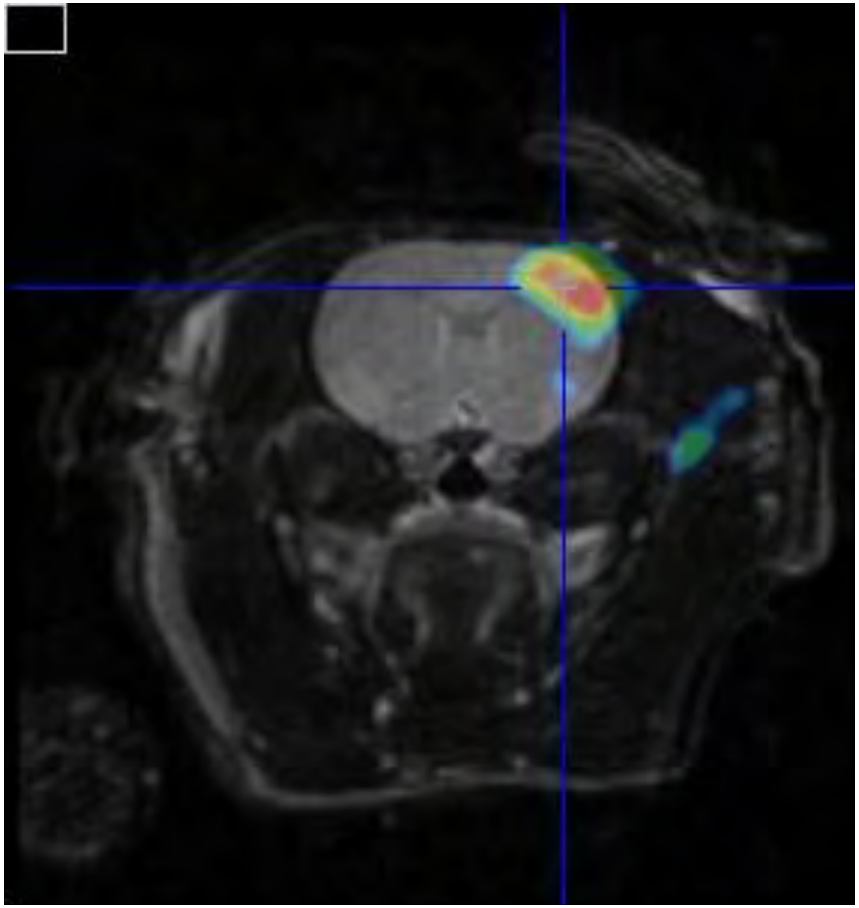
Positron Emission Tomography

^{18}F-FMISO – identification of hypoxic lesions in the brain



^{18}F-FMISO in rats

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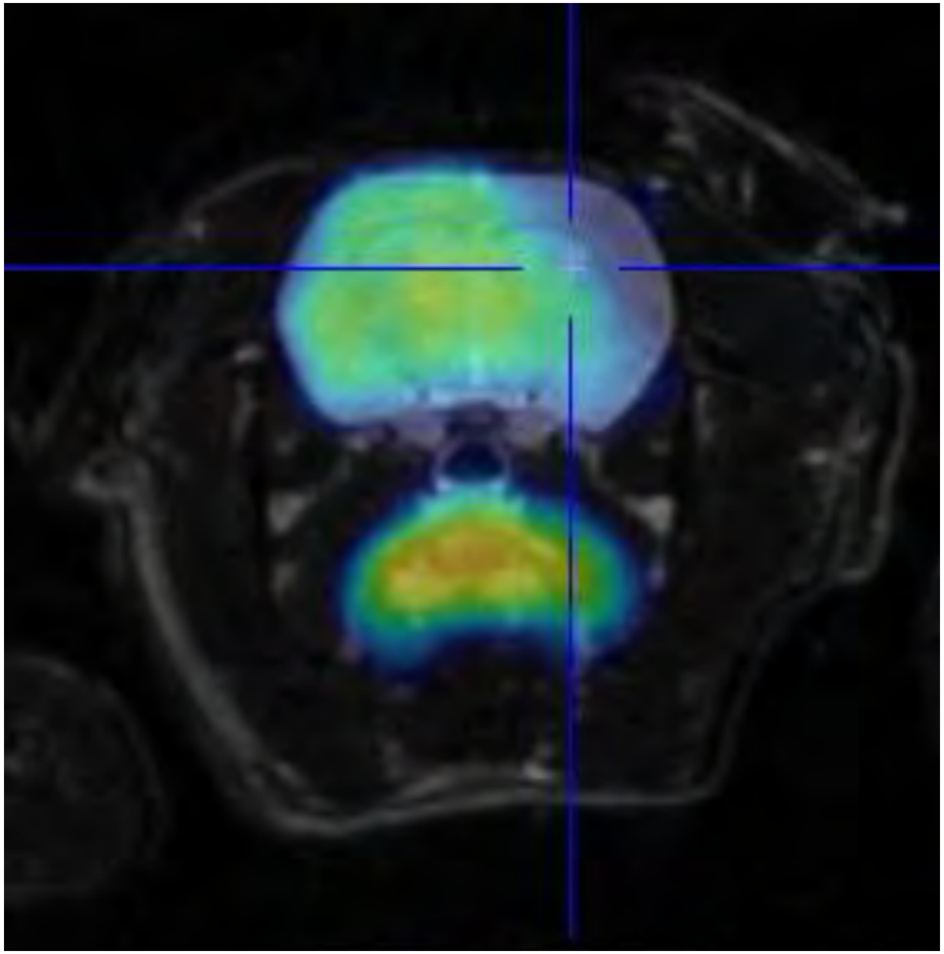
Positron Emission Tomography

^{18}F-FDG Brain Metabolism after Ischemic Injury



^{18}F-FDG in rats

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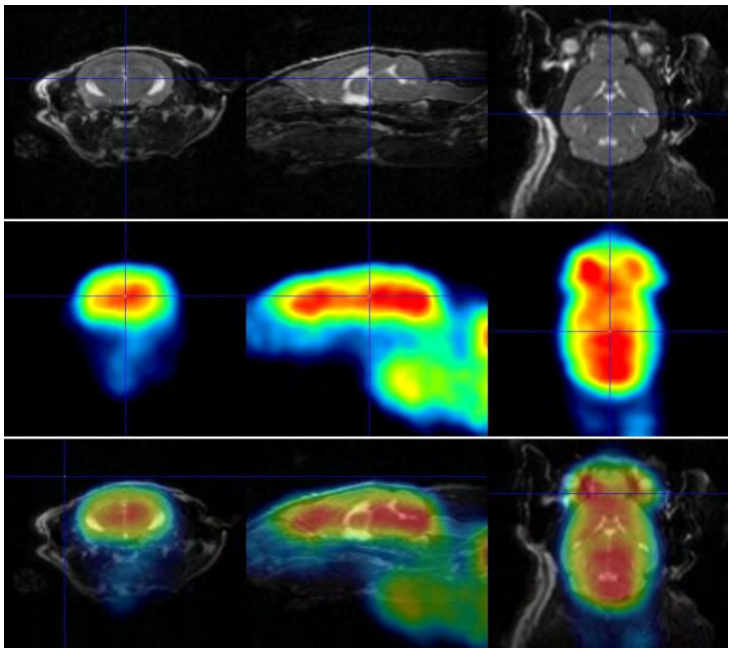
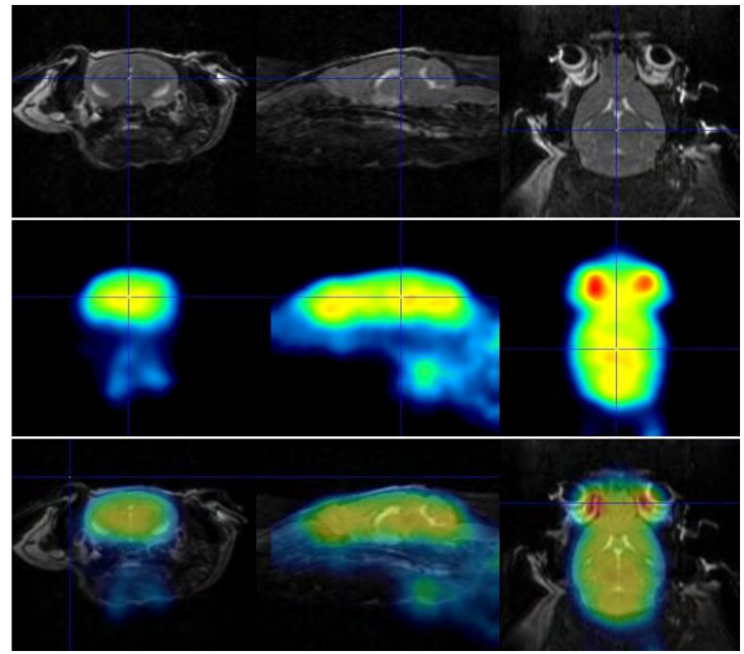
Positron Emission Tomography

Metabolic Changes in Alzheimer´s Models

Control



APP+



Prof. M.A.Pozo Instituto Pluridisciplinar. Universidad Complutense de Madrid

- significant decrease in brain metabolism in mouse models that over express APP
- can be readily visualized with the Albira system and then in this case co-registered with T2 weighted MRI images
- Co-registration done using the supplied PMOD software package and MRI compatible animal transport beds. Animals were iamges with ^{18}F-FDG

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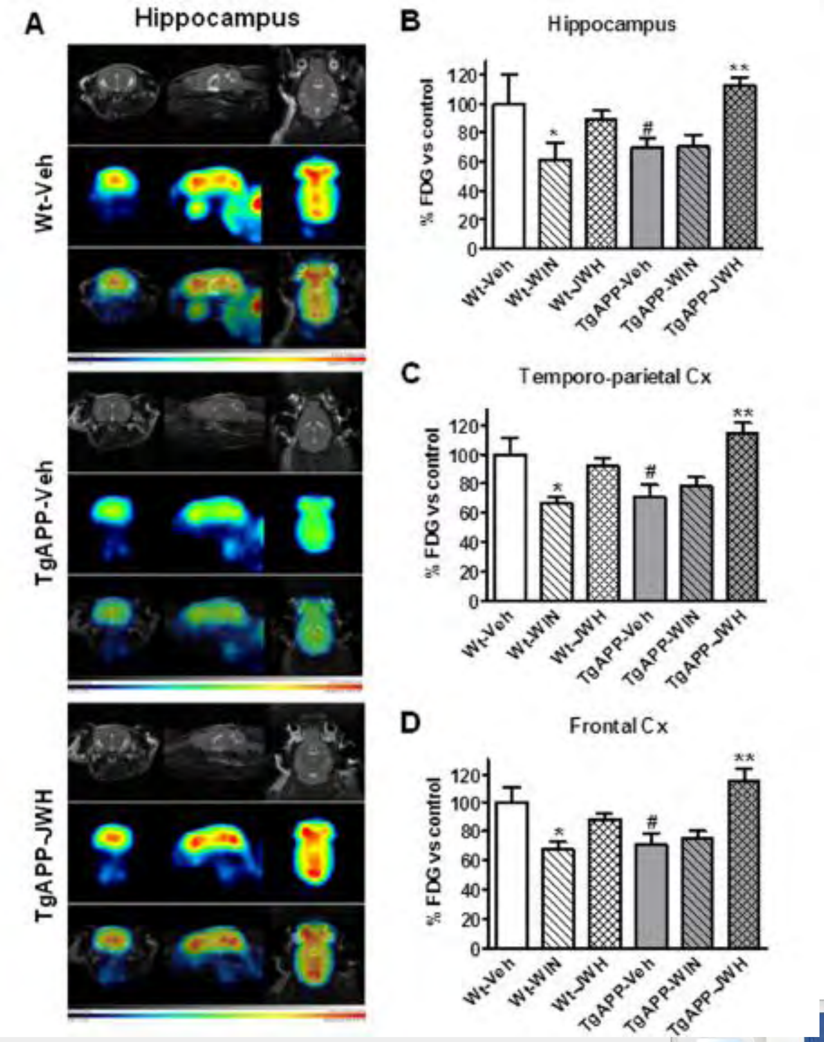
Positron Emission Tomography

Aβ Damage reduced by Cannabinoids – ^{18}F-FDG PET

- App Tg mice show decreased levels of FDG Uptake
- Treatment with CB2 selective agonist JWH rescued glucose metabolism reduction
- CB_2 agonist also rescued behavioral deficits
- Note: CB_1 activation induces psychoactive effects NOT CB_2 activation



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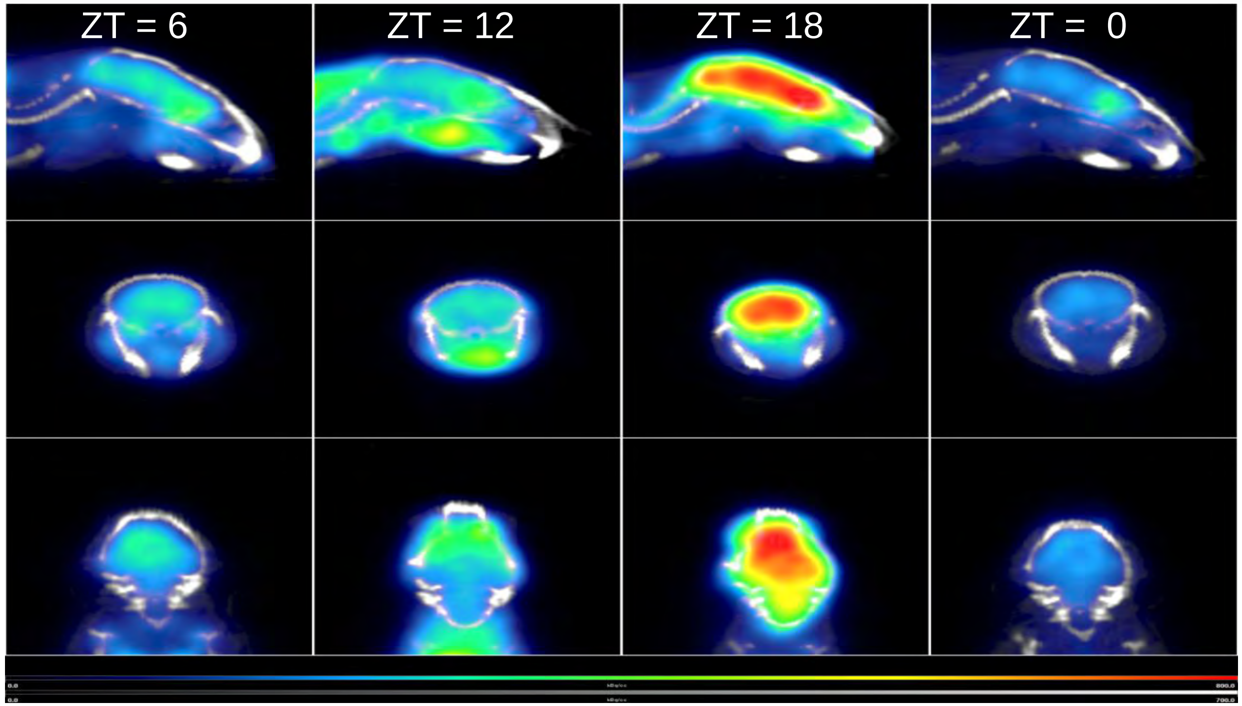
Positron Emission Tomography

Effects of circadian rhythm on brain metabolism – ^{18}F-FDG PET/CT



Dr. Daan van der Veen and graduate student Jinping Shou, from the laboratory of Prof. Giles Duffield, are measuring the effects of circadian rhythms on brain metabolism using *in vivo* PET-CT imaging.

In both whole brain but not in sub brain regions you can observe circadian rhythms in the murine brain with peak activity in the night time. ZT means Zeitgeiber time which is a normalized day night cycle.

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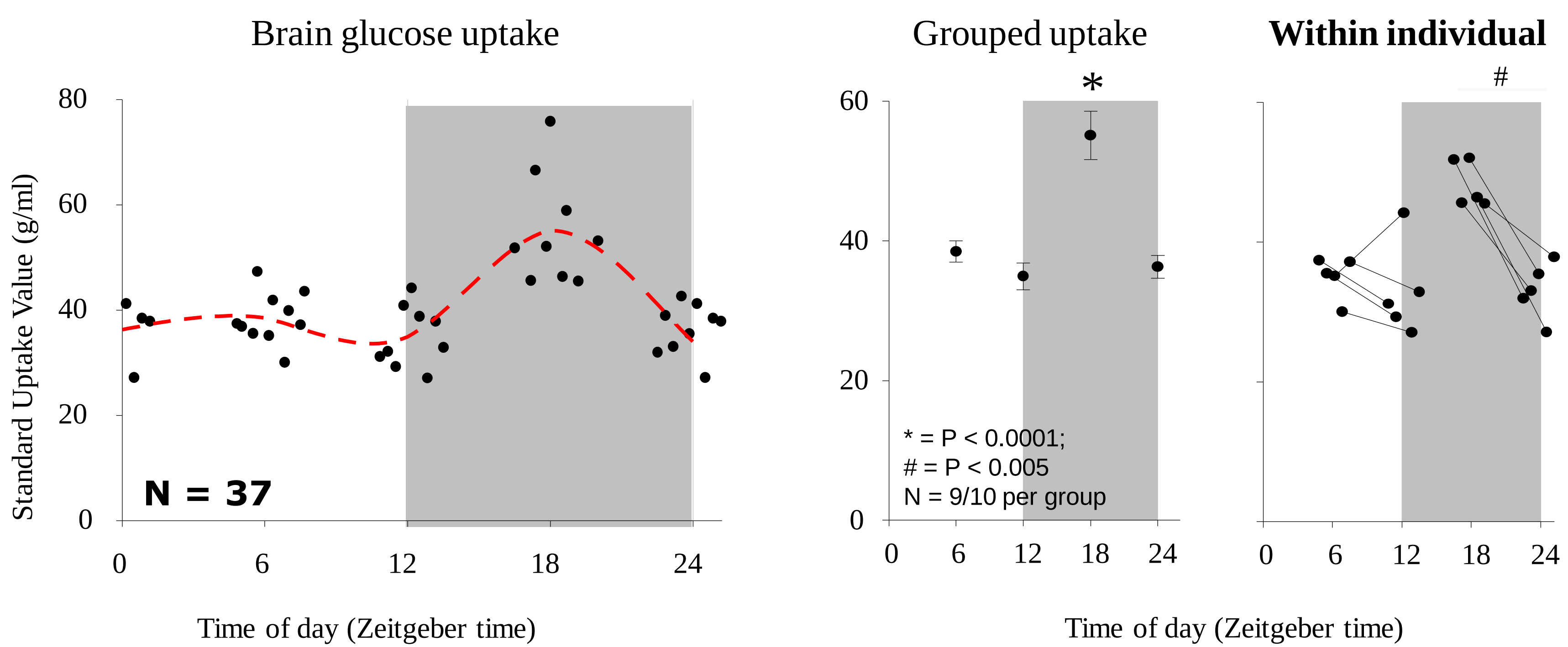
Positron Emission Tomography

Effects of circadian rhythm on brain metabolism – ^{18}F-FDG PET/CT



Van der Veen *et al.*, PLOS One, 2012

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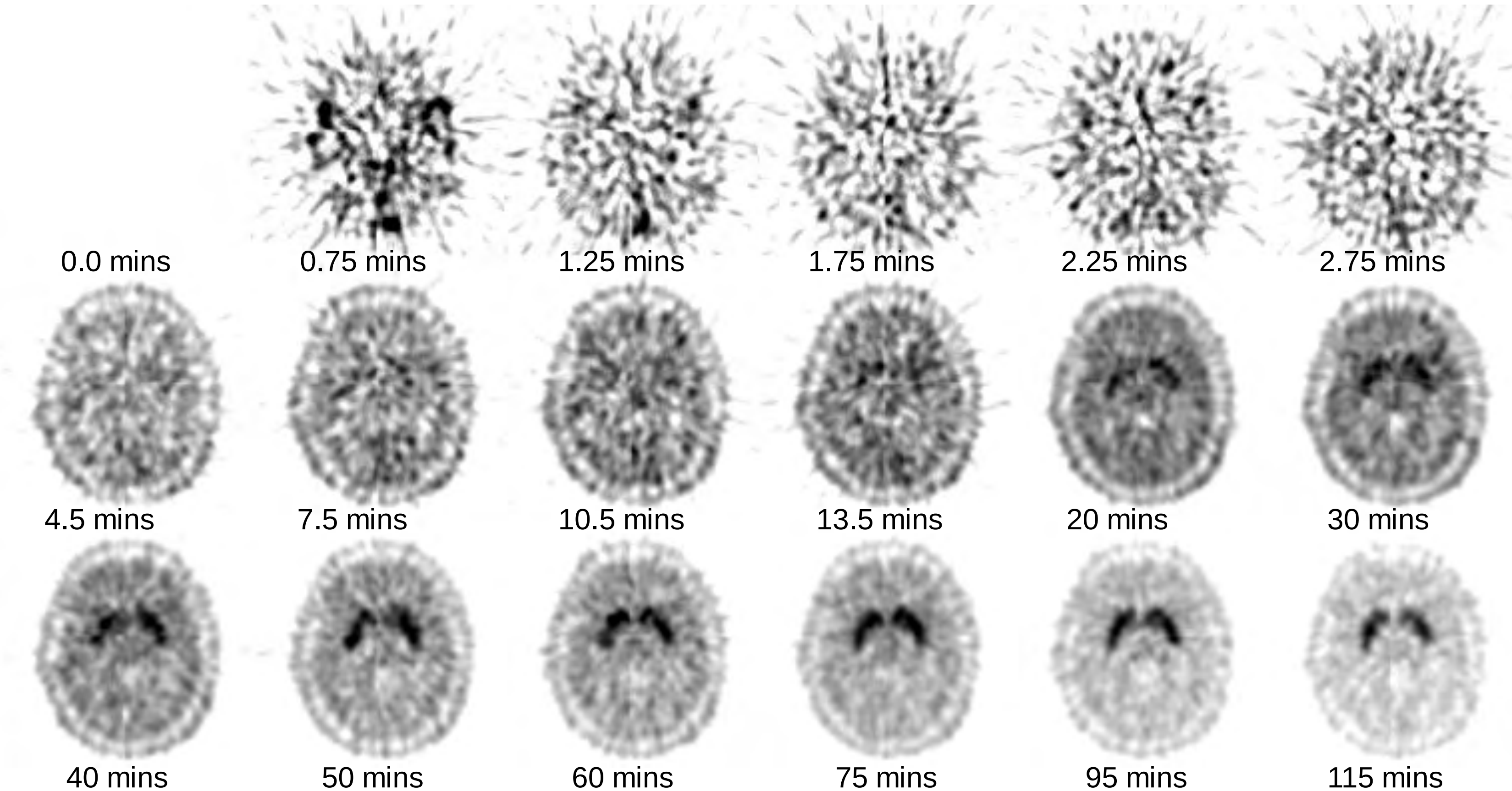
Positron Emission Tomography

^{18}F-FDOPA time course in mouse brain



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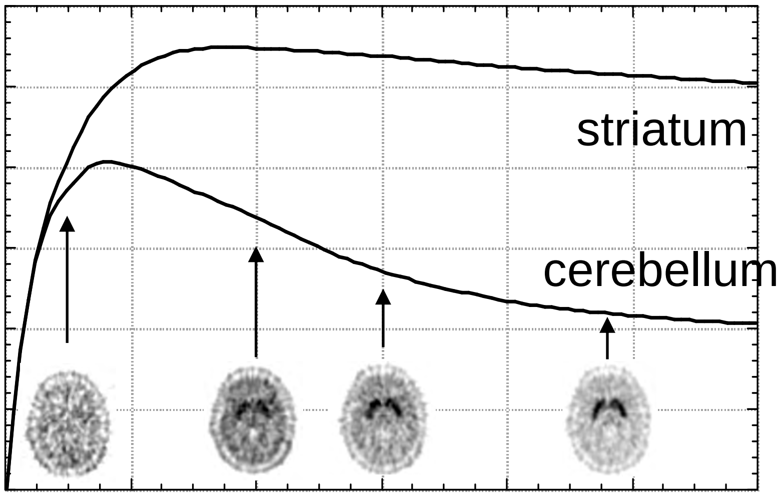
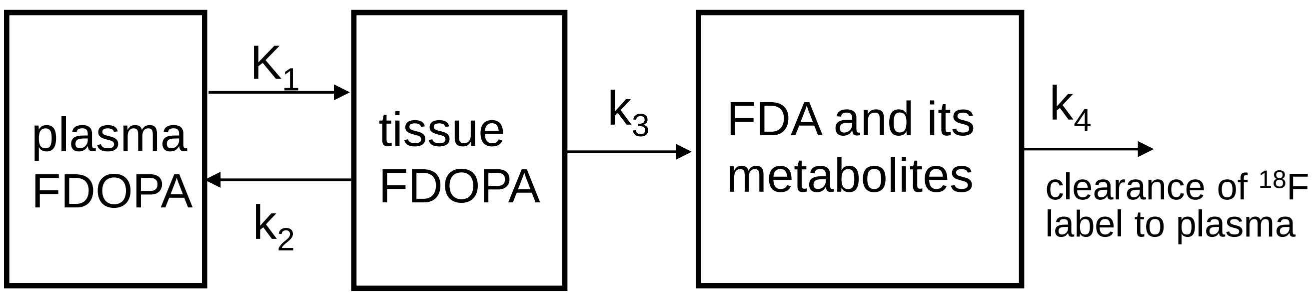
Positron Emission Tomography

^{18}F-FDOPA - Tracer Kinetic Modeling



- Simplified model for FDOPA kinetics in striatum
- Rate constants K_1, k_2, k_3 & k_4 can be estimated using measured PET time activity curves and blood input function.



FDOPA = [^{18}F]fluoroDOPA

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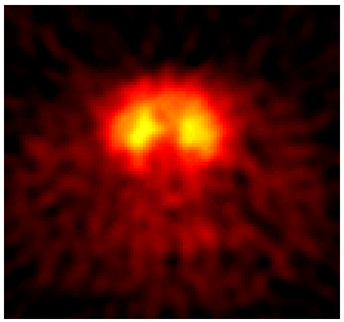
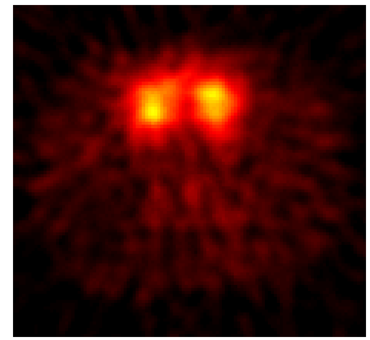
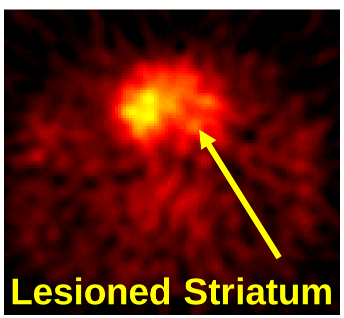
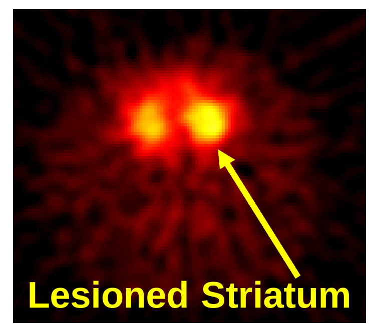
Positron Emission Tomography

Striatal Dopamine System of Rats

[C-11]WIN 35,428 DA Transporter Binding		[C-11]Raclopride DA D2 Receptor Binding
	**Control**	
 	**Unilateral 6-Hydroxydopamine Lesion**	

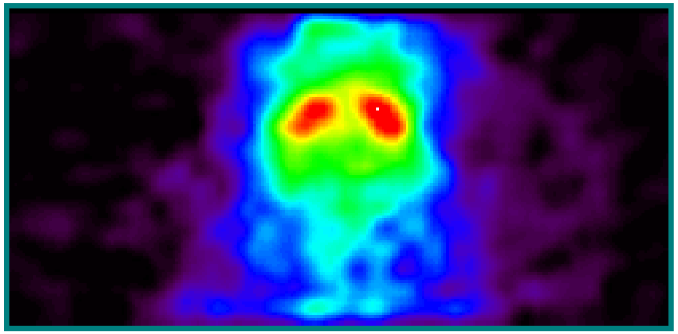
Dan Rubins, Goran Lacan, Simon Cherry, andWilliam Melega

eigner@ujf.cas.cz

Positron Emission Tomography

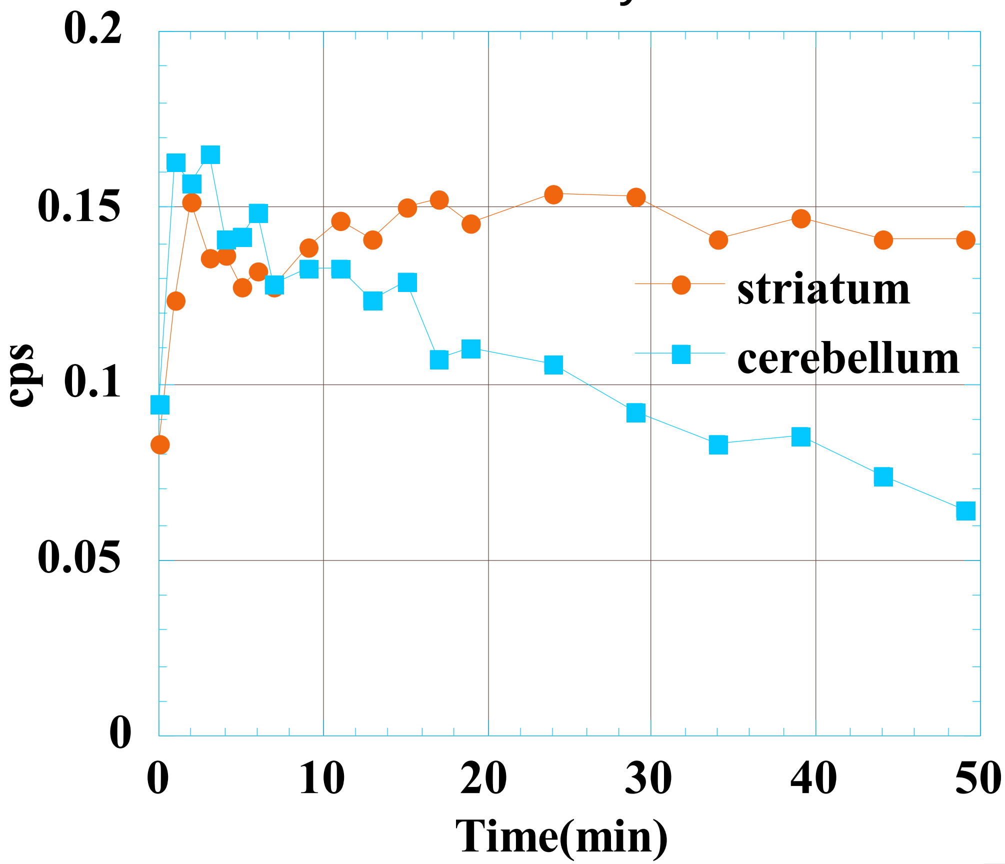
^{11}C-WIN 35, 428 in mouse brain

30g mouse
transverse brain section



180µCi of ^{11}C-WIN 35,428
(0.018µg)

Time Activity curve



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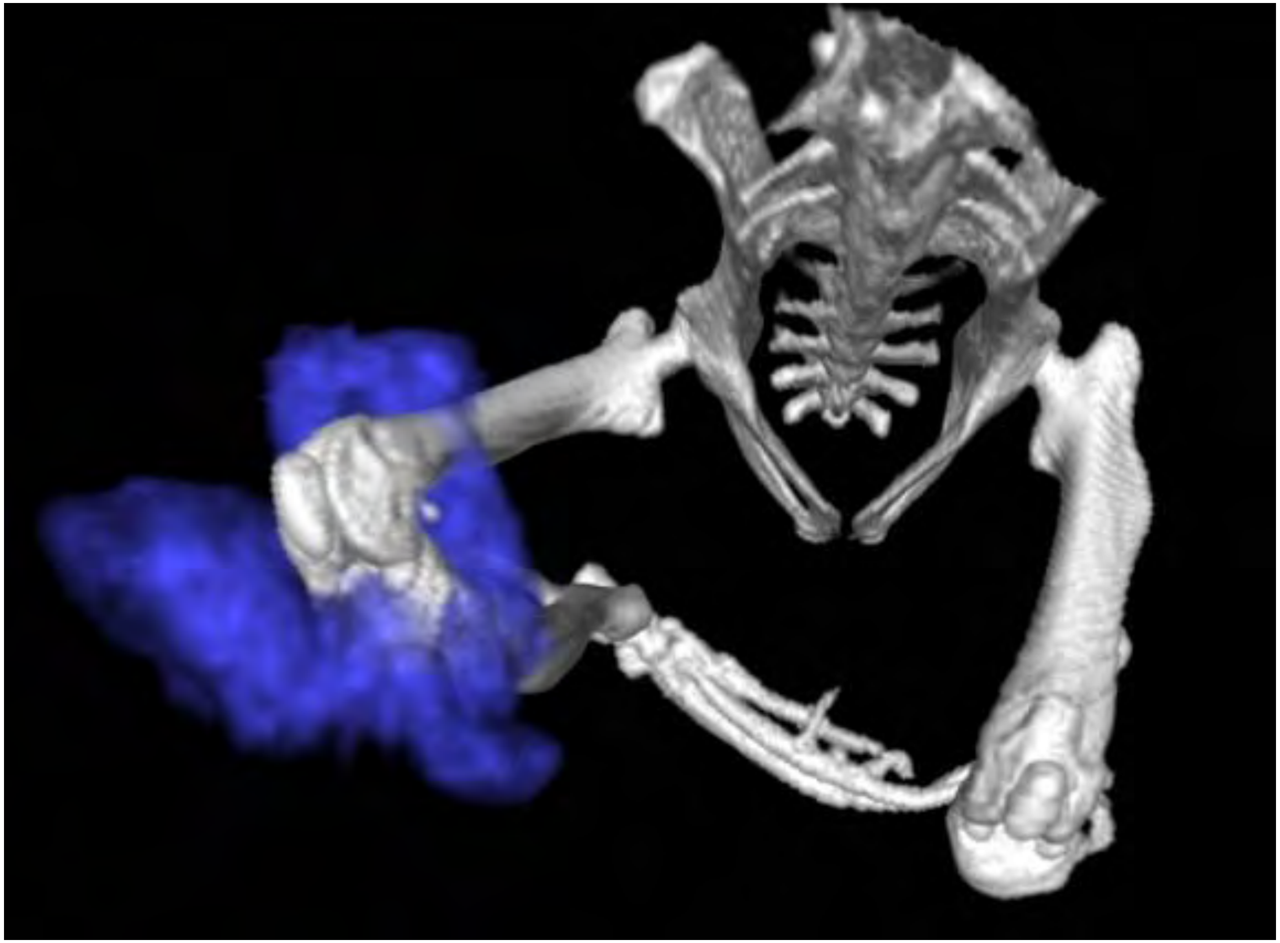
Positron Emission Tomography

Oncology & Therapeutic Evaluation



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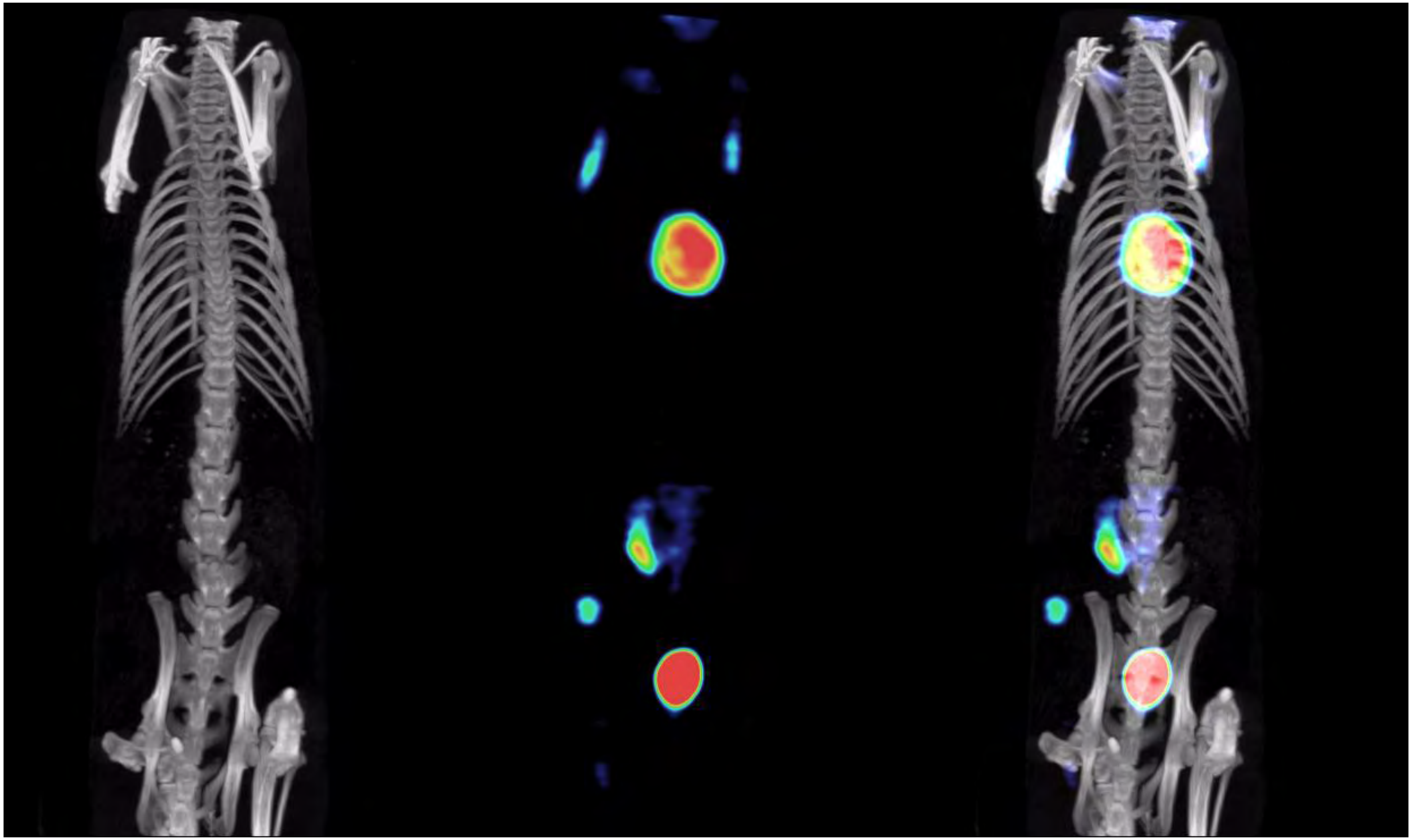
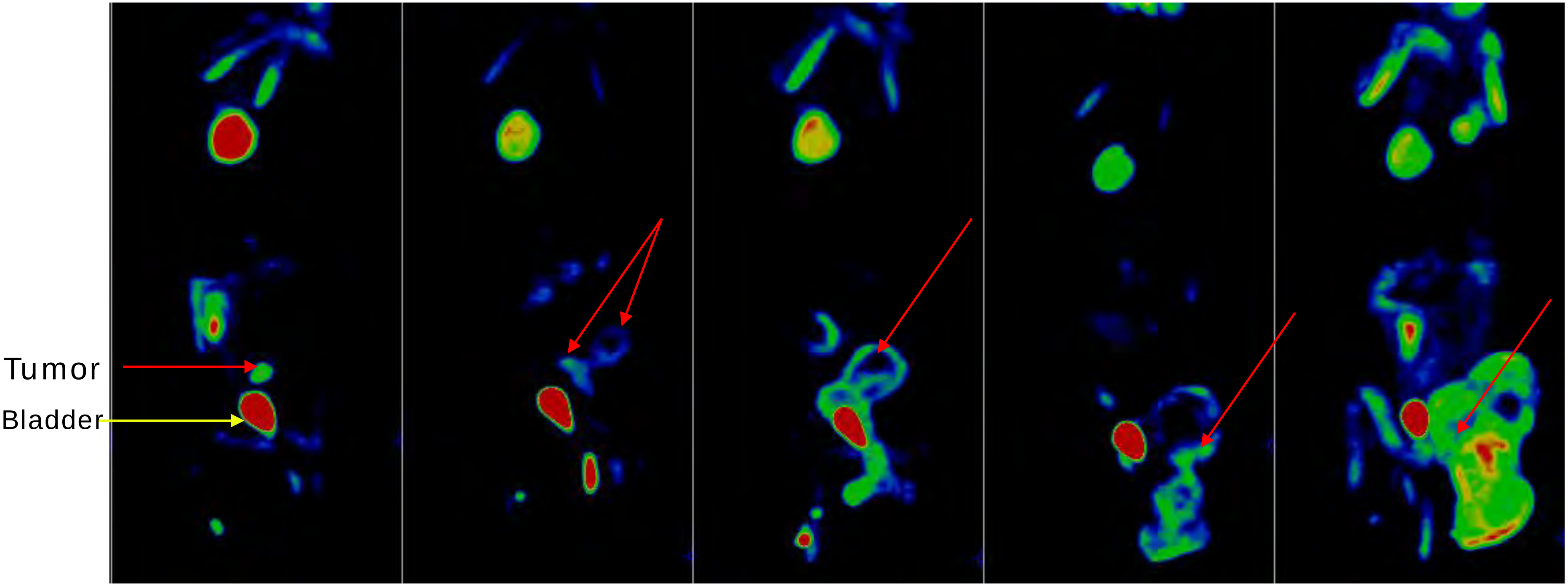
Positron Emission Tomography

Colon Cancer Xenograft – ^{18}F-FDG PET/CT



Positron Emission Tomography

Colon Cancer Xenograft – ^{18}F-FDG PET – 6 week progression



2 weeks 3 weeks 4 weeks 5 weeks 6 weeks

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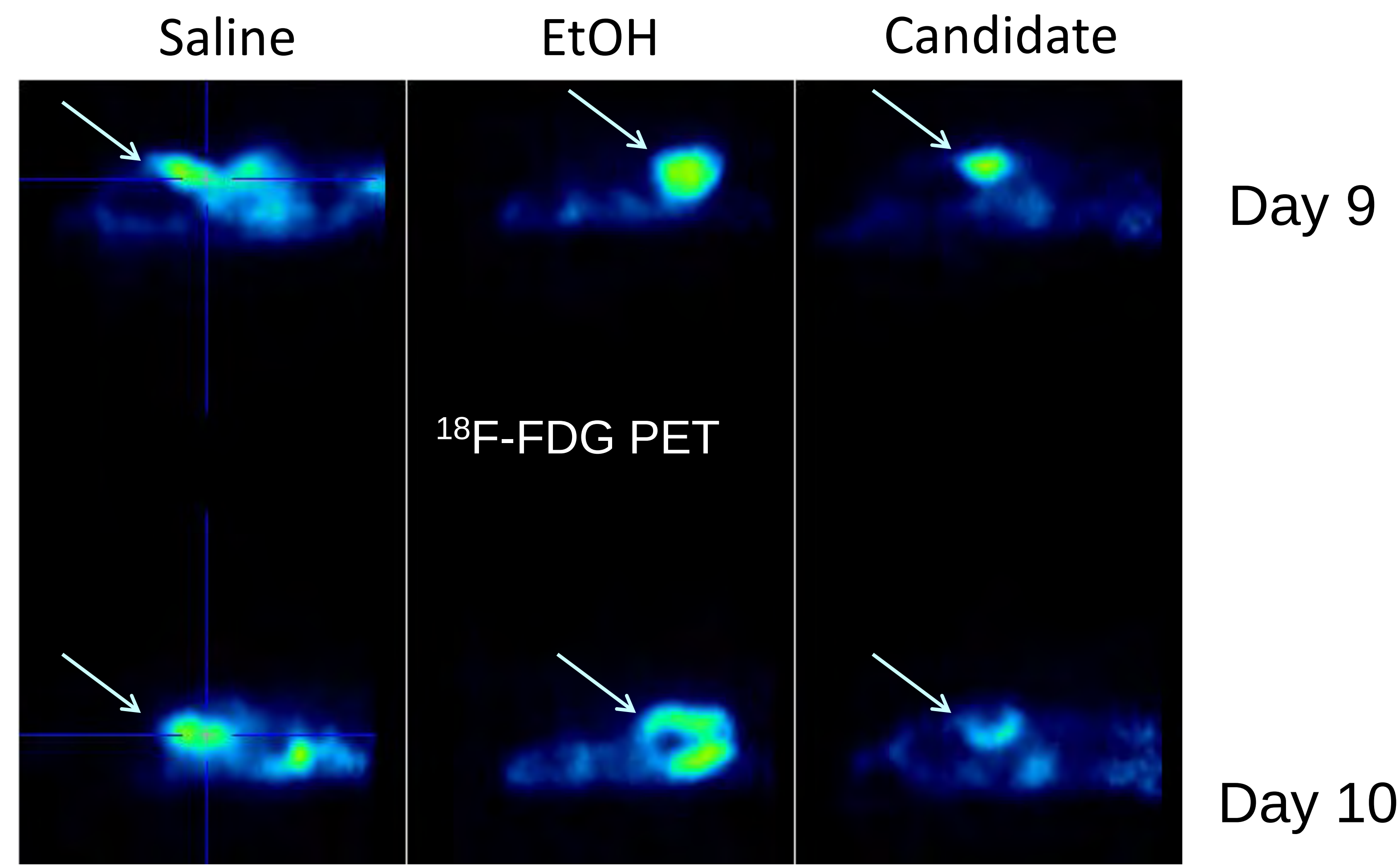
Positron Emission Tomography

Evaluation of locoregional application of chemotherapeutics



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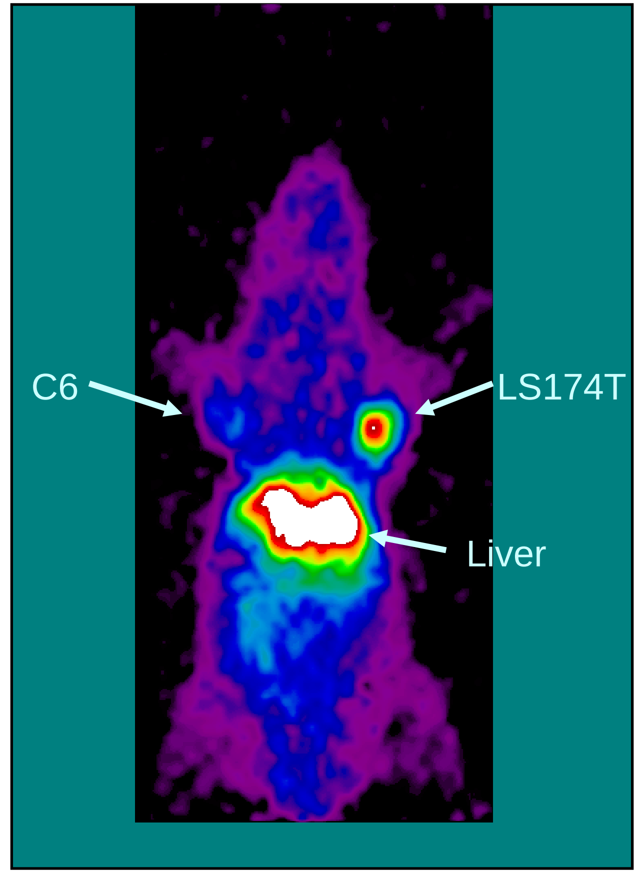
Positron Emission Tomography

^{64}Cu-DOTA anti-CEA Minibody

- Athymic mouse with LS174T (CEA+) and C6 (CEA-) xenografts
- Injected with 70 µCi ^{64}Cu-anti-CEA minibody (engineered antibody fragment, scFv-C_{H3})
- Scanned 12 hr post injection
- Courtesy of Anna Wu (UCLA and City of Hope)



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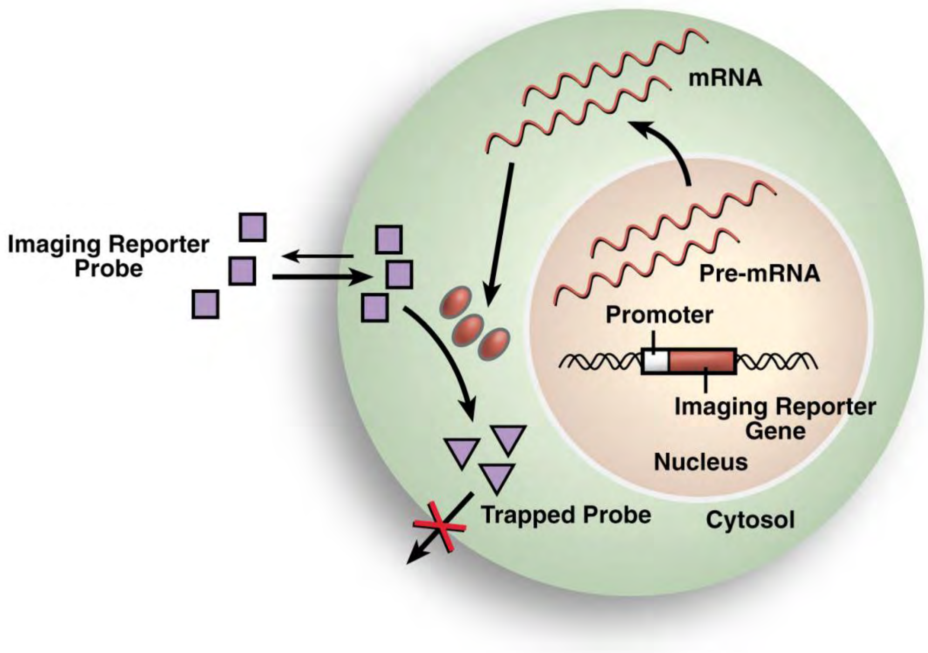
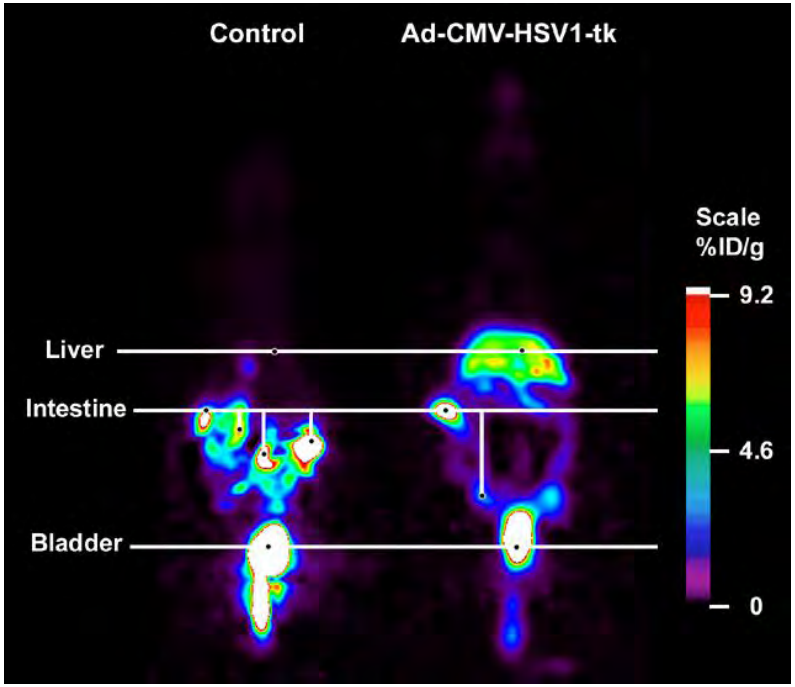
Positron Emission Tomography

Imaging gene Expression by PET





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Positron Emission Tomography

microPET in Drug Development

- direct radiolabeling of drug
  - biodistribution and pharmacokinetics
- binding/competition studies
  - dosing and pharmacodynamics
- indirect markers
  - pharmacodynamic effect on secondary marker (e.g. metabolism or blood flow)

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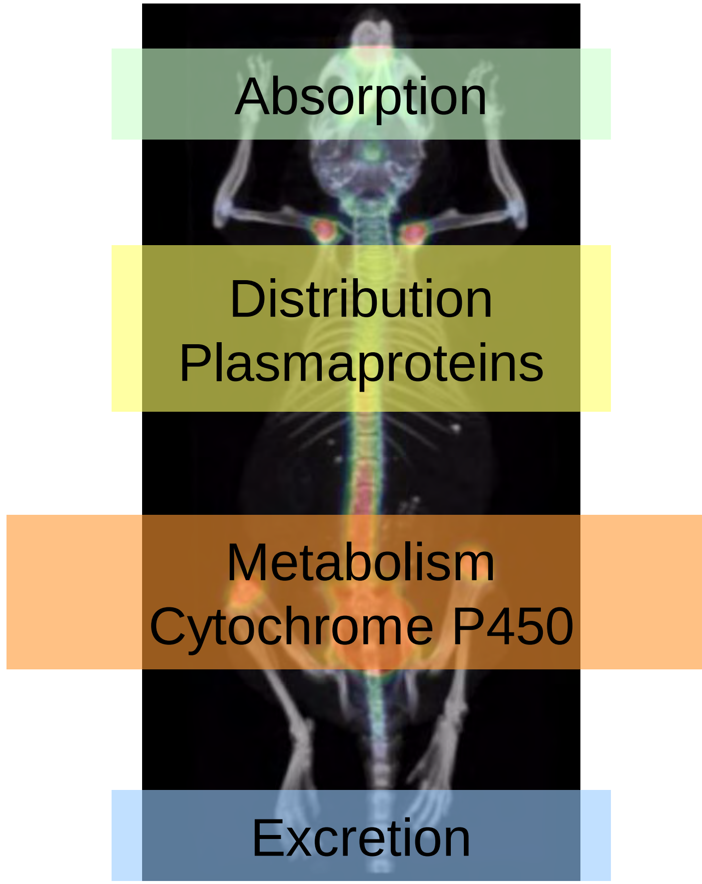
Pharmacokinetic

Absorption; Distribution, Metabolism, Excretion



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Radiopharmacy

Routine Production, Custom Synthesis & Labeling Concepts

Nuclear Physics Institute of the Academy of Sciences of the Czech Republic, p.r.i.
Department of Radiopharmaceuticals

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Dr. Katerina Eigner Henke

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Thank You

for Your Attention

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