


TEHRAN UNIVERSITY  
OF  
MEDICAL SCIENCES



TUMS PRECLINICAL  
CORE FACILITY (TPCF)



## FLUORESCENCE PLANAR MOLECULAR IMAGING

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Associate Professor of TUMS  
CEO of TANP

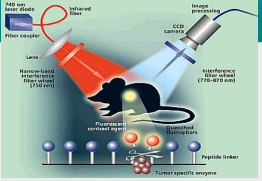
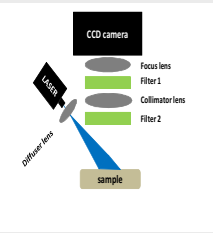
TPCF Workshop May 28<sup>th</sup>-29<sup>th</sup> 2019, Manila, Philippines

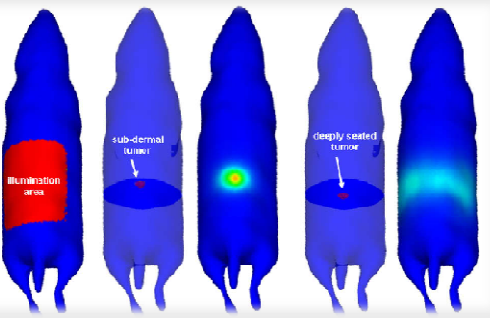


- “The visual representation , characterization and quantification of biological processes at cellular and subcellular levels within intact living organisms.”(Massoud and Ghambir ,2003)
- Combining the targeting technology of molecular biology with the detection technology of imaging instrumentation to image and monitor both cellular and animal physiology and function in-vivo

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
## Instrumentation



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## Instrumentation

- There are characteristics of Fluorescence Small Animal Imaging
  - In vivo  $\neq$  in vitro
  - Value of integrating molecular events with cellular and animal physiology in vivo for basic biological research
  - Can efficiently survey whole animals
  - Potential for **rapid in vivo screening**
  - Eventually bridge between animal studies and human studies.

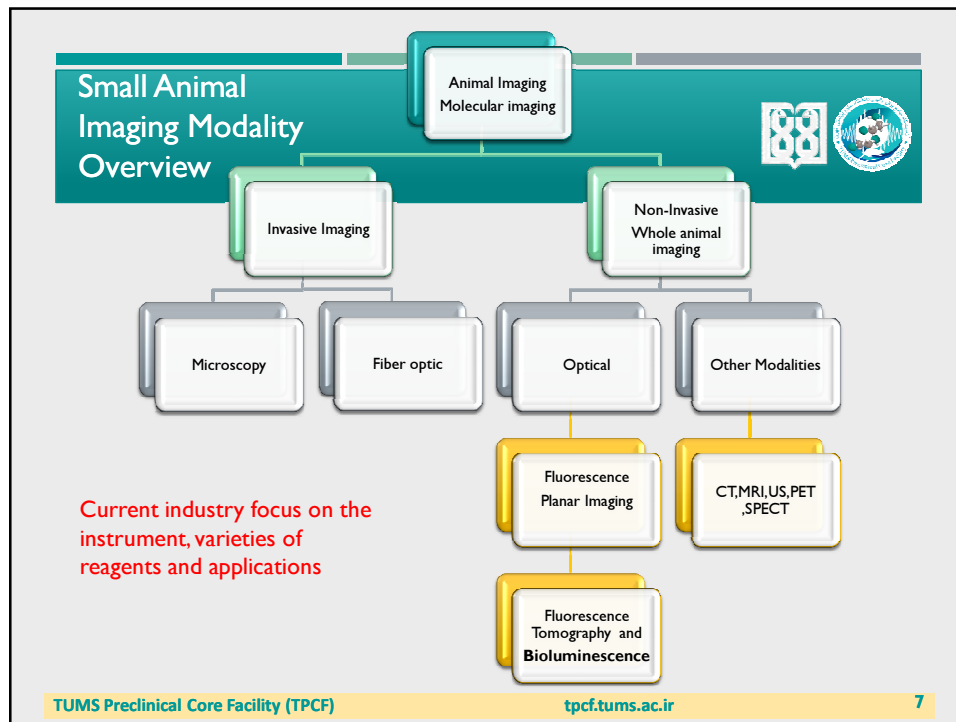
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**Pharmaceuticals**  
**Pharmacology**  
**Immunology**  
**Stem cell research**  
**Neuroscience**  
**Oncology**  
**Small animal imaging**

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| target expression  | drug bio-distribution and PK  | drug-target interaction   | signal transduction   | cellular physiological & metabolic effect  | effects on structure & micro-structure                                     |
|--|---|---|---|--|--|
| is the target expression specific for the organ?<br>is the target expression altered during pathology? | is the drug absorbed & distributed?<br>how fast is the drug eliminated?<br>is the drug reaching its target tissue in sufficient amount? | is the drug reaching its molecular target?<br>what is the receptor occupancy of the drug? | is the drug-target interaction promoting response at molecular level? | is therapeutic intervention promoting biological response at physiological or metabolic level (system response)? | is therapeutic intervention causing macroscopic structural changes/repair? |

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### Comparison of Small Animal In Vivo Imaging

- High Sensitivity, low cost & ease of use take optical Imaging to the advance modality
- Combining Imaging modalities Enables Animal Physiology in light of anatomy

| Modality  | Anatomy | Animal Physiology | Metabolism | Molecular | Cellular Physiology | Cost | Sensitivity | Depth      | Resolution  |
|---|---------|-------------------|------------|-----------|---------------------|------|-------------|------------|-------------|
| US<br>Ultrasound                                    | High    | Low               | Low        | Low       | Low                 | Low  | High        | mm         | 50 μm       |
| CT<br>X-ray Computed Tomography                     | High    | Low               | Low        | Low       | Low                 | High | Low         | No Limit   | 50 μm       |
| MRI<br>Magnetic Resonance Imaging                   | High    | Low               | Low        | Low       | Low                 | High | Low         | No Limit   | 10 - 100 μm |
| PET<br>Positron Emission Tomography                 | Low     | High              | High       | High      | High                | High | High        | No Limit   | 1 - 2 mm    |
| SPECT<br>Single Photon Emission Computed Tomography | Low     | High              | High       | High      | High                | High | High        | No Limit   | 1 - 2 mm    |
| BLI<br>Bioluminescence Imaging                      | Low     | High              | High       | High      | High                | Low  | High        | cm         | Several mm  |
| FT<br>Fluorescence Tomography                       | Low     | High              | High       | High      | High                | Low  | High        | 5 - 6 cm   | 1 - 2 mm    |
| FI<br>Fluorescence Imaging (Planar)                 | Low     | High              | High       | High      | High                | Low  | High        | < 1 cm     | 1 - 2 mm    |
| Confocal Microscopy                                 | Low     | Low               | Low        | Low       | Low                 | Low  | High        | 0 - 150 μm | 2.5 μm      |

Adapted from Weissleder (2002) Nature Reviews Cancer 2:1-8

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**Optical Imaging At Benchtop**

**Compound prescreening**

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**OBJECTIVES**

Where

Figure 3. External image of bone metastasis of AC3488-GFP. External images of tumours in the skeletal system including (a) the skull, (b) scapula, and (c) spine in a dorsal view of live intact nude mouse.\*

Hoffman, Robert M. "Green fluorescent protein imaging of tumour growth, metastasis, and angiogenesis in mouse models." *The lancet oncology* 3.9 (2002): 546-556.

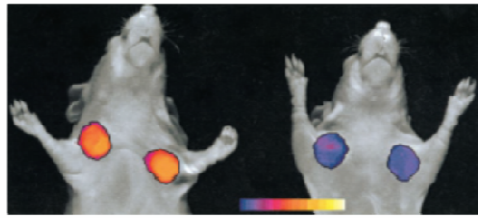
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## OBJECTIVES



### Why Functional Activity

Fig. 3. Real-time imaging of protease inhibition. Control mouse (left) and treated mouse (right), both with two H1-1000 tumors (overexpressing MMP-2). Tumors from the mouse pretreated with an MMP-2 inhibitor show markedly decreased fluorescence compared with untreated tumors (from Ref. 30, reprinted with permission).

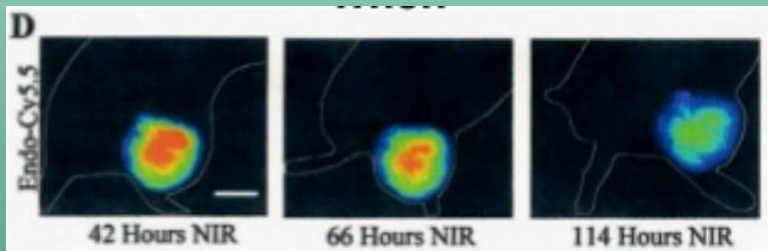


Mahmood, Umar, and Ralph Weissleder. "Near-infrared optical imaging of proteases in cancer." *Molecular cancer therapeutics* 2.5 (2003): 489-496.

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### When Near Infrared images after injection with endostatin-Cy5.5

Goal:  
High  
Content  
In Vivo  
Imaging



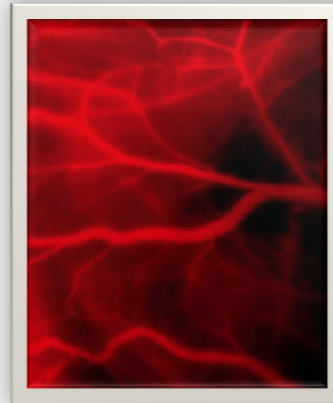
Hassan, Moinuddin, and Brenda A. Klaunberg. "Biomedical applications of fluorescence imaging in vivo." *Comparative medicine* 54.6 (2004): 635-644.

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## In Vivo vascular imaging



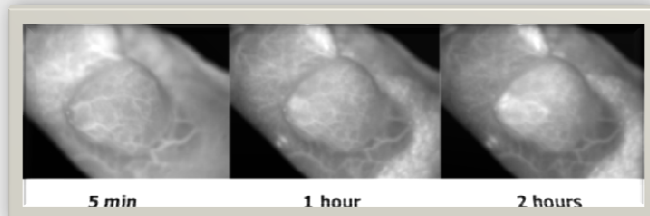
- Venous injection at increasing resolution
- Bright signals allows highly detailed vascular analysis
- Red colors allow deeper, higher resolution
- Long circulation times allows detailed vascular imaging



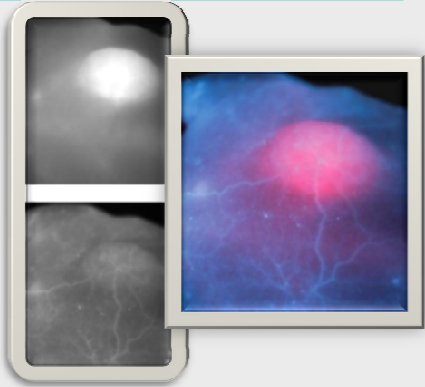
## Monitoring Tumor Blood Physiology



- Non-targeted quantum dot

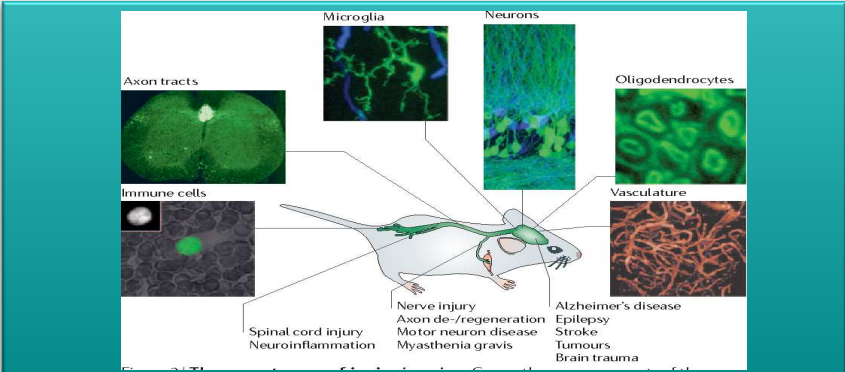


Multiplex and composite



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Brain Imaging



Misgeld, Thomas, and Martin Kerschensteiner. "Neuroimaging: In vivo imaging of the diseased nervous system." *Nature Reviews Neuroscience* 7.6 (2006): 449.

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## TANP Company




FluoVision is a range of systems for in vivo fluorescence imaging. It provides real time images and videos of fluorescent signals in living animals, for non-invasive imaging, surgery or dissection (intravital imaging), for small and large animals.

**Main Indications**

- Oncology
- Biodistribution & targeting probe development
- Lymph nodes
- Cardiovascular research
- Immunology
- Infectious diseases

**Benefits**

- High sensitivity
- Great flexibility with the open space design
- Works with white light
- Records in real time images and videos
- Easy to install, to use, to move

Tajhiz Afarinan Noori Parse Co.





**FluoVision<sup>®</sup>  
Preclinical  
System**

Tajhiz Afarinan Noori Parse Co.

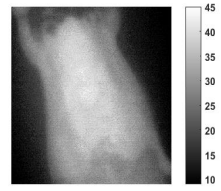
Tel: +98 21 6690 7527  
Fax: +98 21 6658 1533  
www.tanp.ir  
info@fluovision.com

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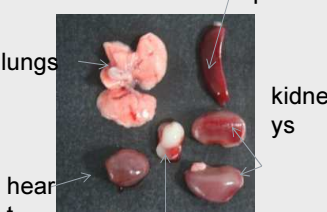
## Optical imaging Applications: Imaging Gene Expression

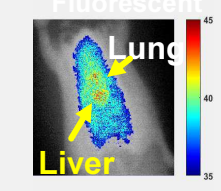
- Step 1:** Sepsis animal model, Male C57BL/6 mice
- Step 2:** 10<sup>6</sup> GFP-labeled cells were injected to mice
- Step 3:** GFP - will fluoresce when excited by light
- Step 4:** 24 hours after cell injection Image the mouse



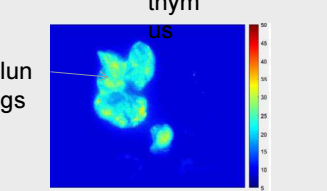
Fluorescent



spleen  
lungs  
kidneys  
heart  
thymus



Lung  
Liver





lungs

Ref: TPCF, Optical imaging laboratory, 2017


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## Optical Imaging Applications: Drug Delivery

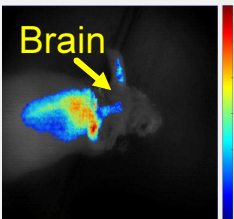



- ❑ **Step 1:** no difference in animal model of interest
- ❑ **Step 2:** drug for Alzheimer disease label to FITC
- ❑ **Step 3:** Inject the drug to the mouse/Rat
- ❑ **Step 4:** Trace the drug with optical imaging method

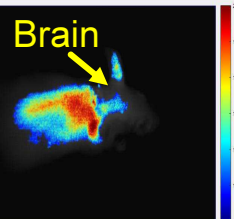
**Bef**



**After 30**



**After 60**



Ref: TPCF, Optical imaging laboratory, 2017  
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## GFP MOUSE

