ÉCOLE POLYTECHNIQUE
FÉDÉRALE DE LAUSANNE

Non-Invasive Imaging of Metabolic Fluxes and Enzymatic Activity in Living Mice

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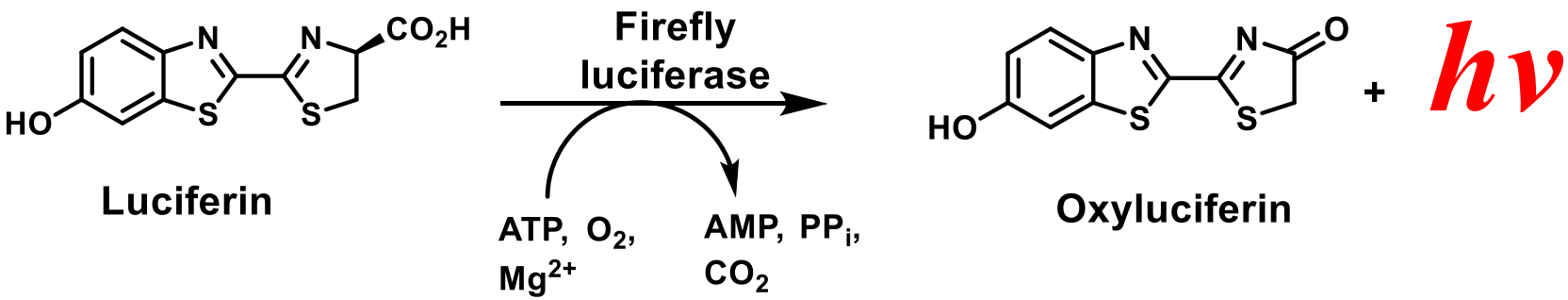
June-6-14

Laboratory of Bioorganic Chemistry and Molecular Imaging (LCBIM)

Development of Probes and Methods for *in vitro* and *in vivo* Imaging

- A novel approach for imaging metabolic fluxes *in vivo*
- A novel approach for imaging of proteases *in vivo*

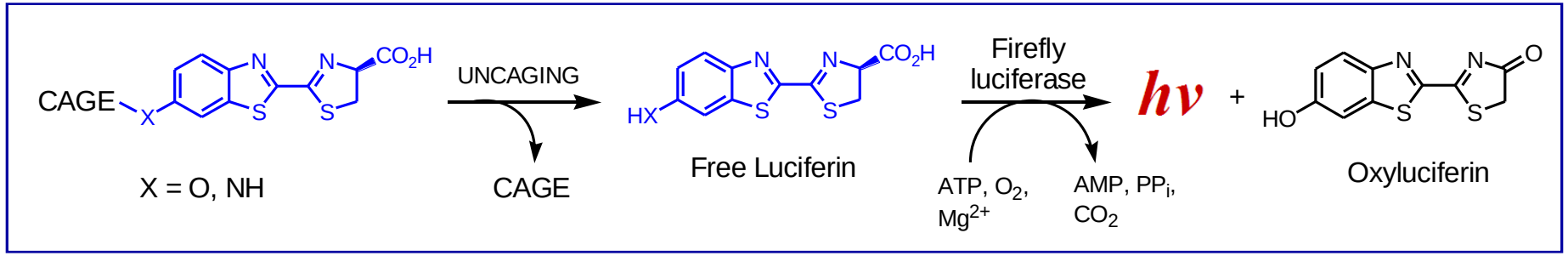
Bioluminescent imaging (BLI)



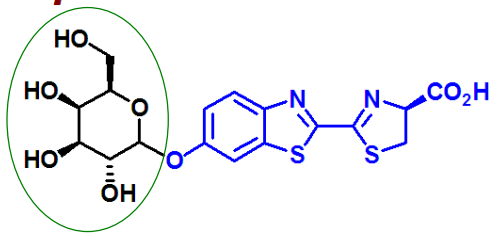
Natural process based on the oxidation of D-luciferin by Luciferase

- Non-invasive detection method
- High sensitivity 10^{-17} mol/L
- 100,000 times more sensitive than fluorescence
- High specificity
- Low background

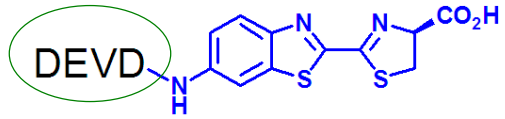
Applications of BLI to probe molecular signatures of target tissues



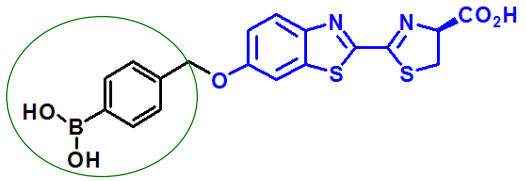
β-Galactosidase [1]



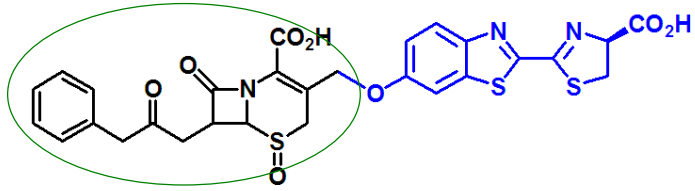
Caspase 3 [2]



H_2O_2 [3]



β-Lactamases [4]

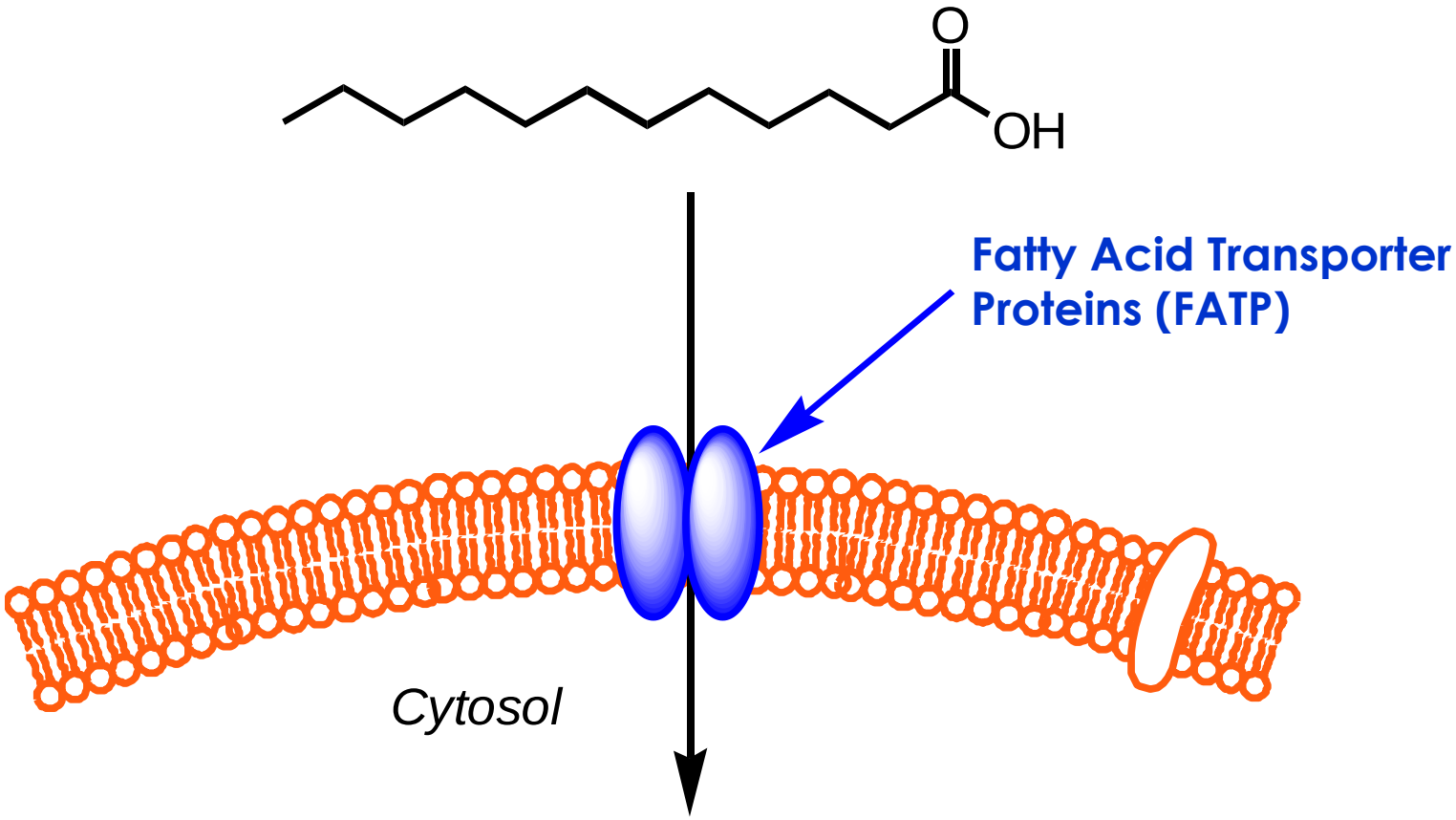


(1) *Nat Methods*, **2006**, *3*, 295. **(2)** *Mol Ther*, **2005**, *11*, 926. **(3)** *Proc. Natl. Acad. Sci.* **2010**, **(4)** *Angew. Chem. Int. Ed.* **2007**, *46*, 7031.

A novel approach for imaging metabolic fluxes in vivo

with Prof. Andreas Stahl Group
Nutritional Science and Toxicology Department,
UC Berkeley, CA

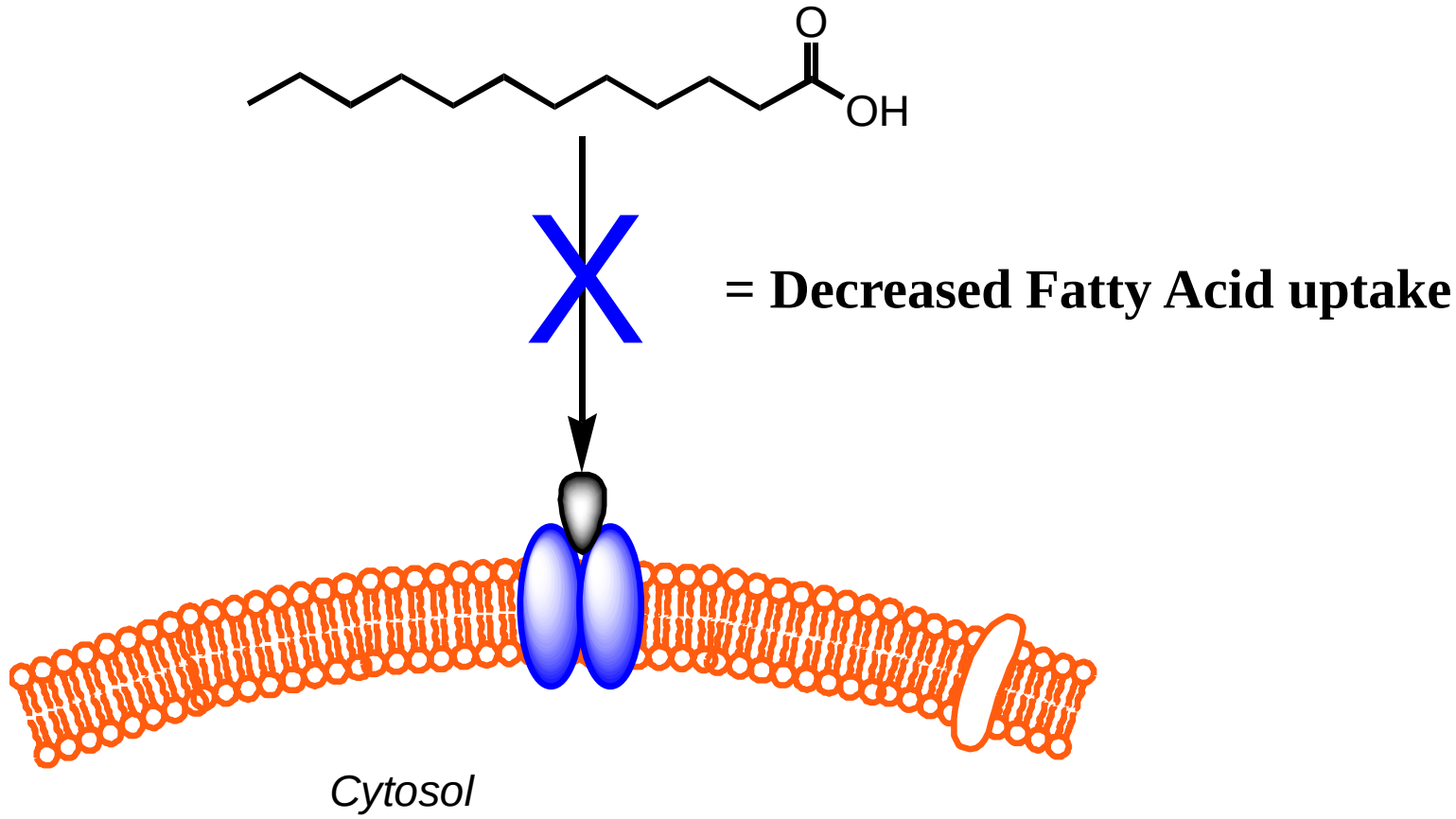
Uptake of fatty acids into cells



FFA uptake regulated by FATP
FATP linked to obesity

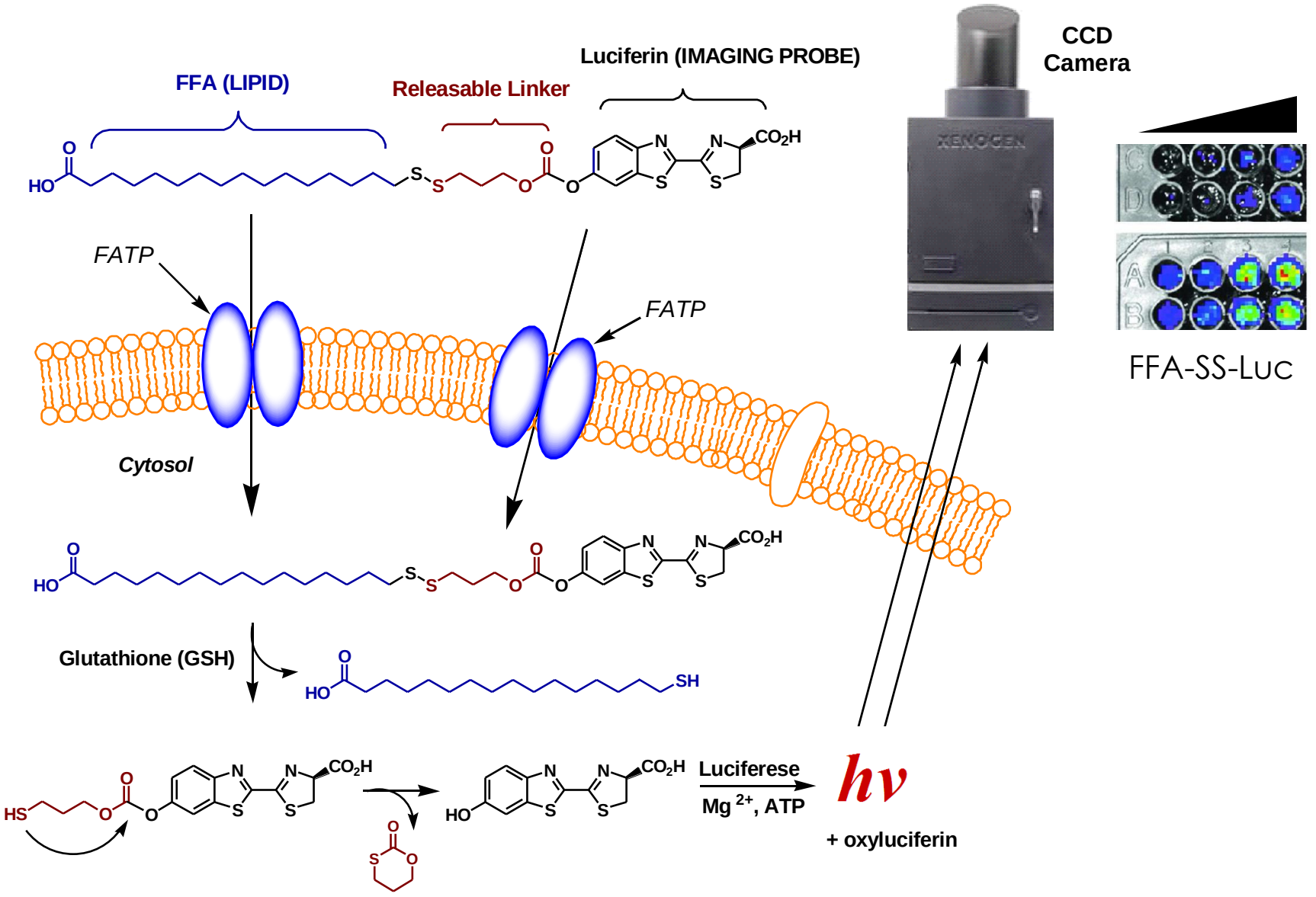
Pflugers Arch. (**2004**) *447*, 722

Blocking FATP in the intestine reduces caloric uptake

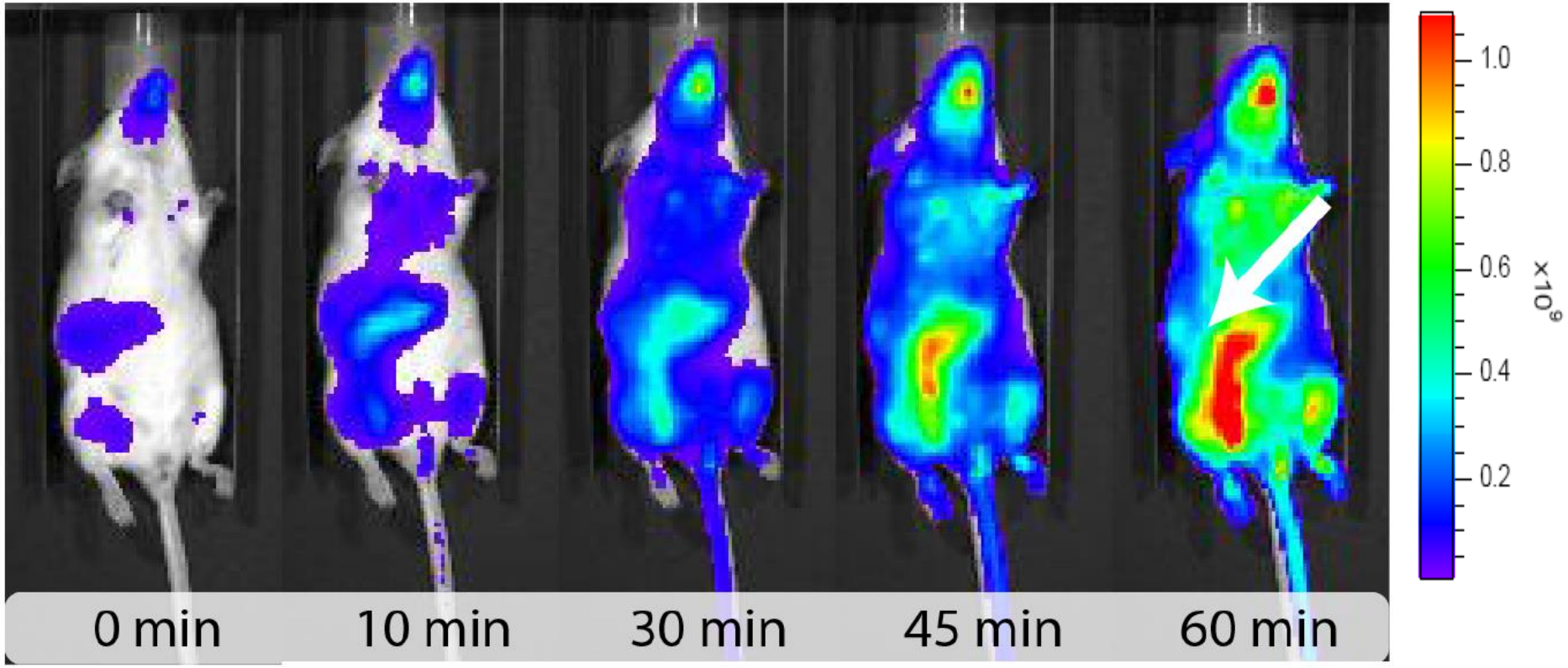


FATP promising therapeutic target to control FFA uptake

Activable bioluminescent fatty acid probe



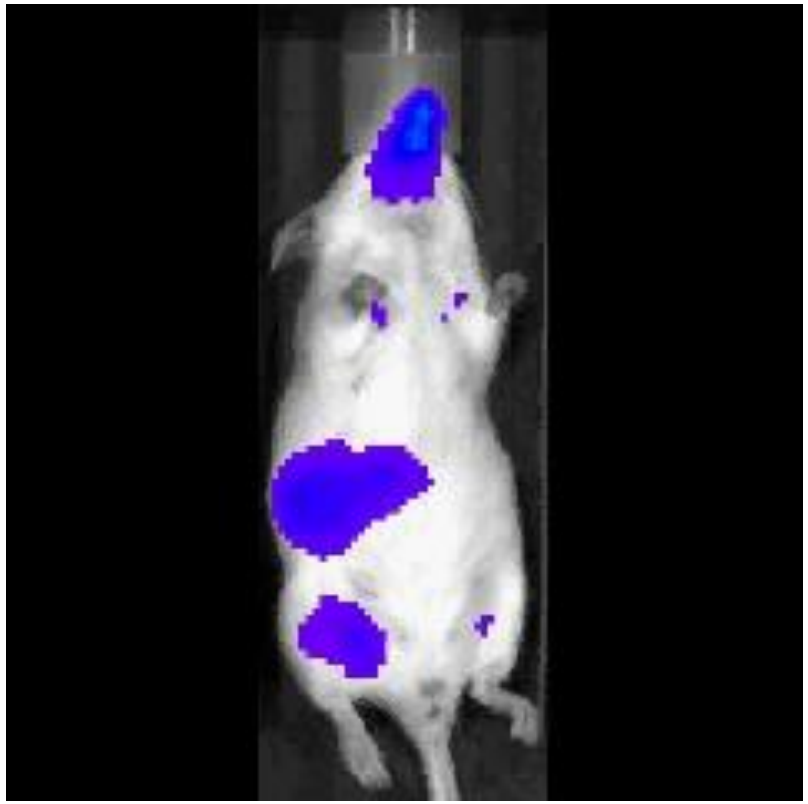
Intestinal fatty acid uptake in real time in live animals



Time course after oral gavage

ACS Chem Biol, **2012**, *7*, 1884

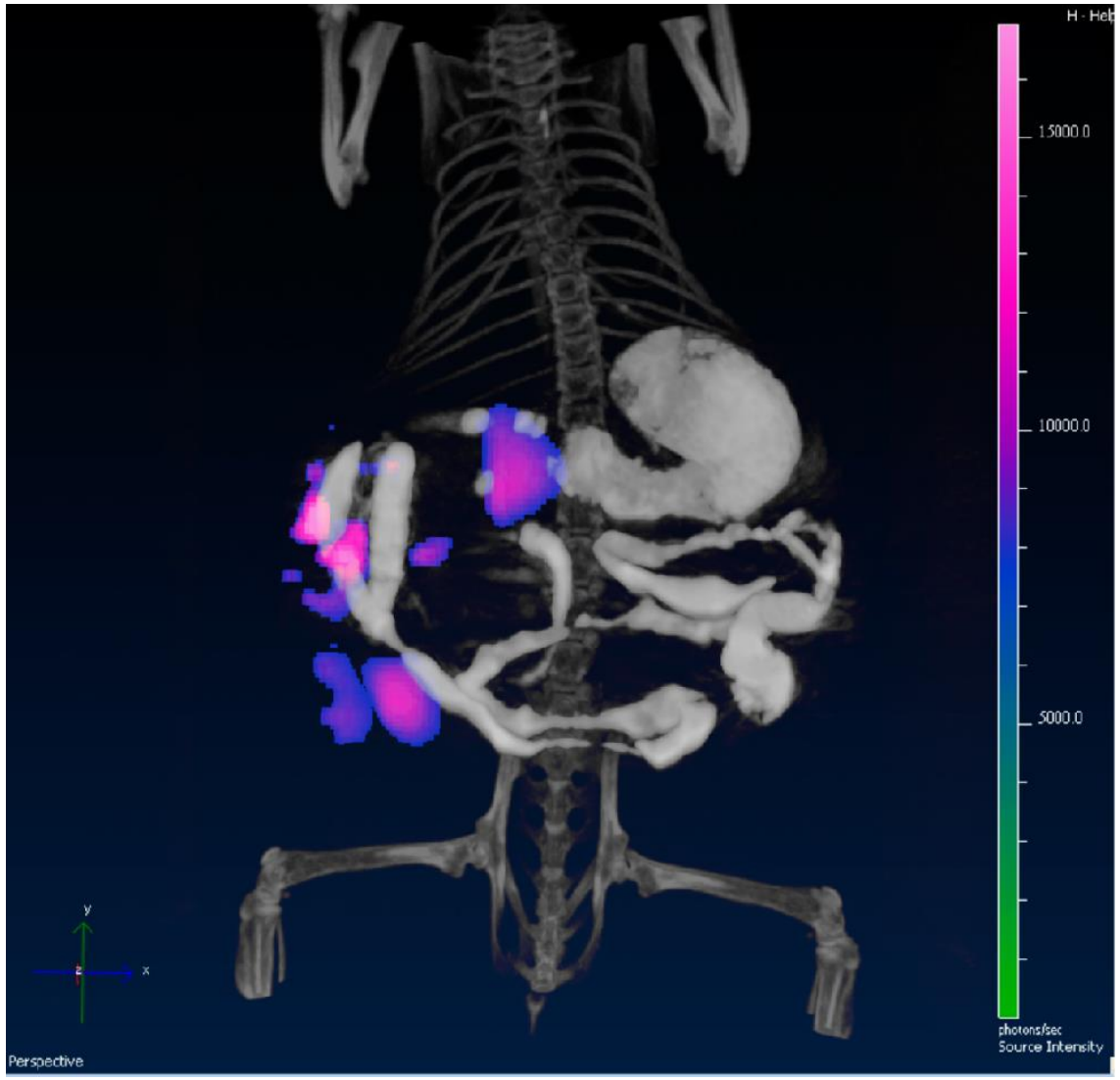
Intestinal fatty acid uptake in real time in live animals



ACS Chem Biol, **2012**, *7,* 1884

MicroCT and BLI overlay of uptake

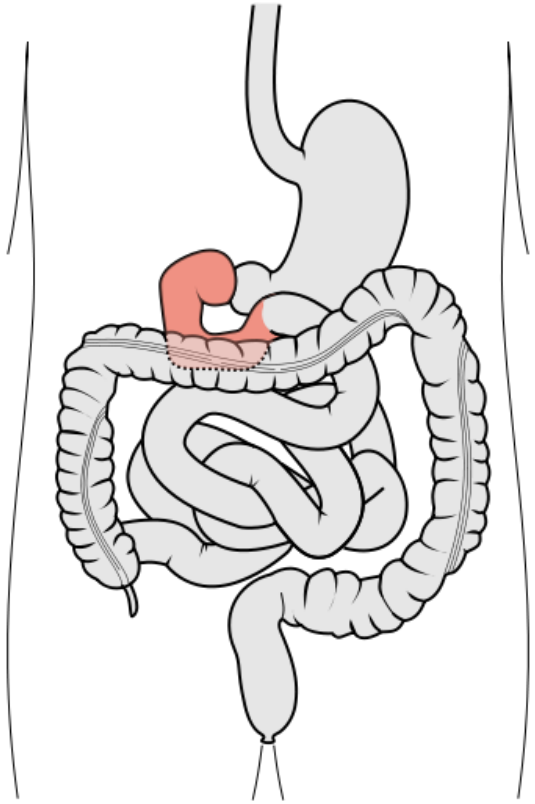
$BaSO_4$ radiocontrast agent for intestine labeling

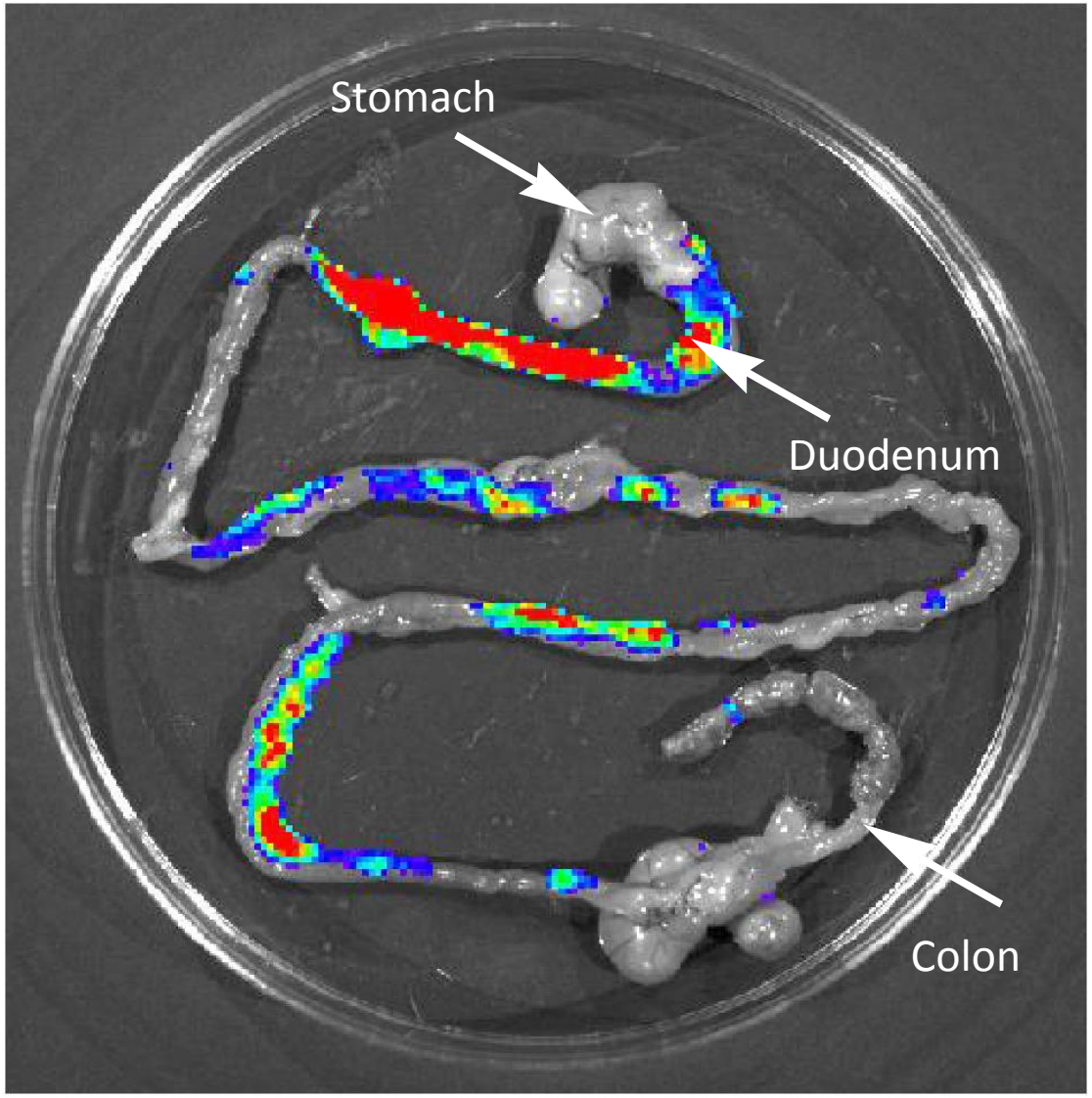


ACS Chem Biol, **2012**, *7,* 1884

Where does the light come from?

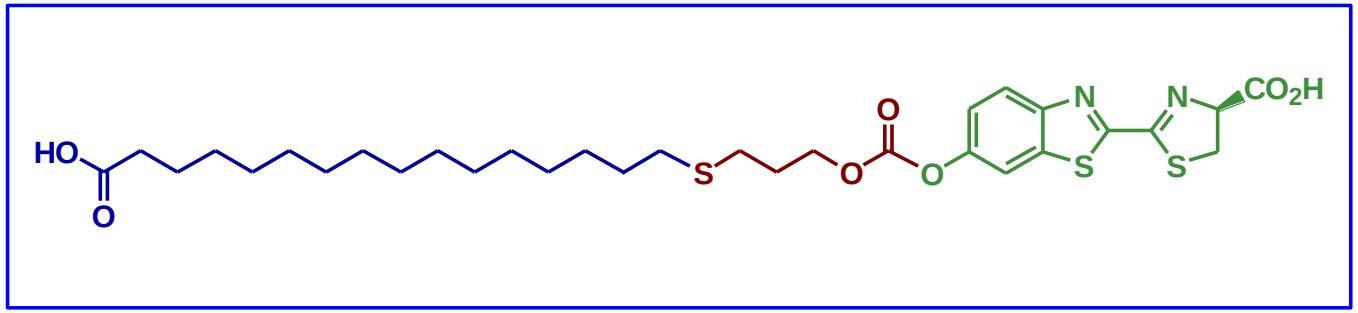
The duodenum is the first section of the small intestine

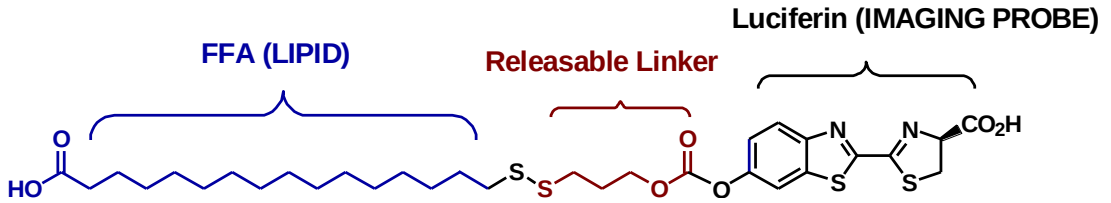


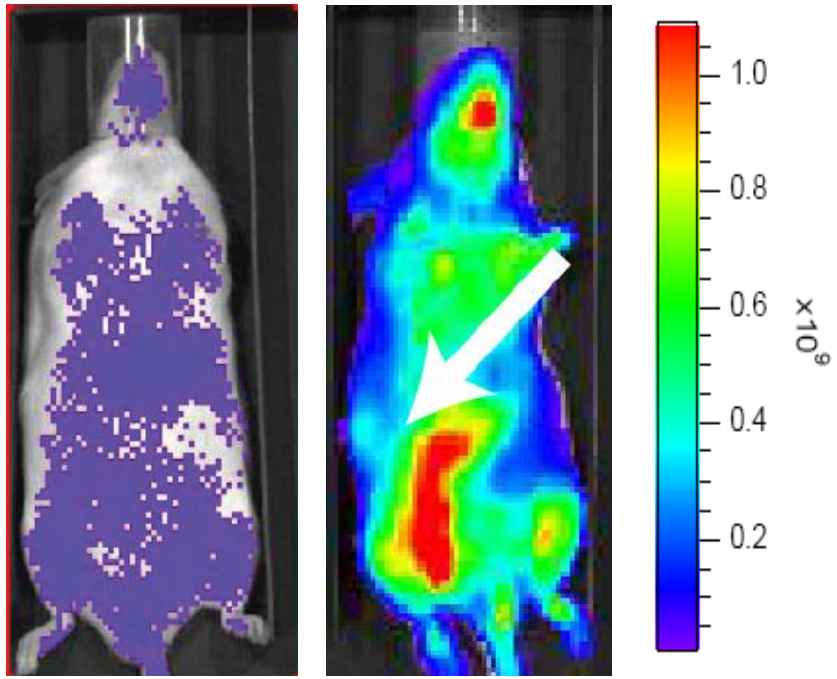


ACS Chem Biol, **2012**, *7,* 1884

Comparison to the control compound



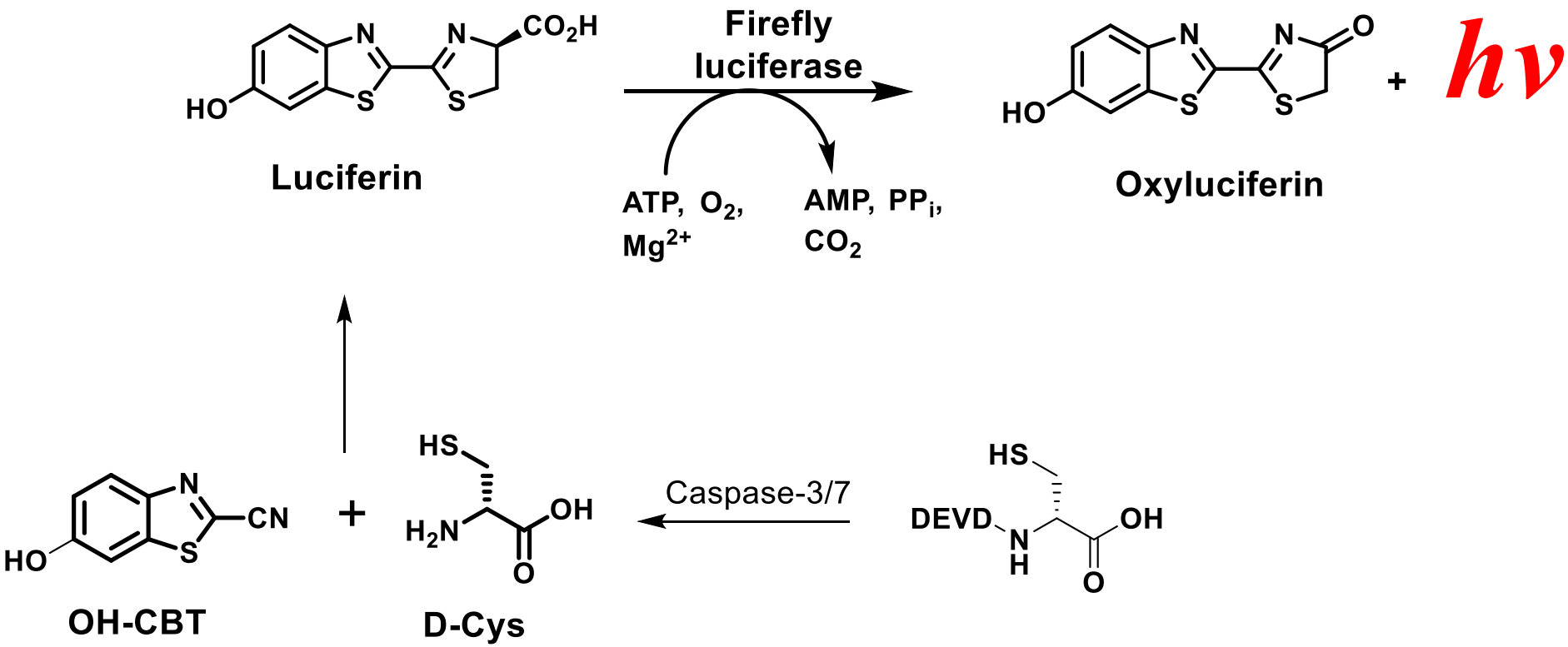




A novel approach for imaging of proteases *in vivo*

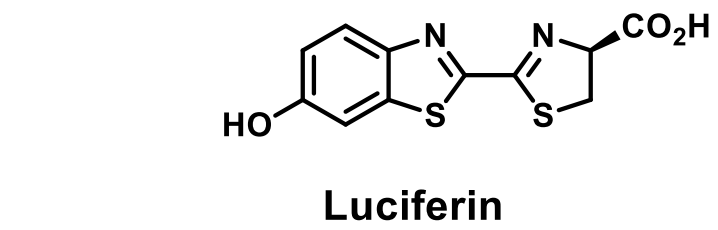
Godinat et. al, ACS Chem. Biol., **2013**, *8* (5), 987–999

Split Luciferin Approach

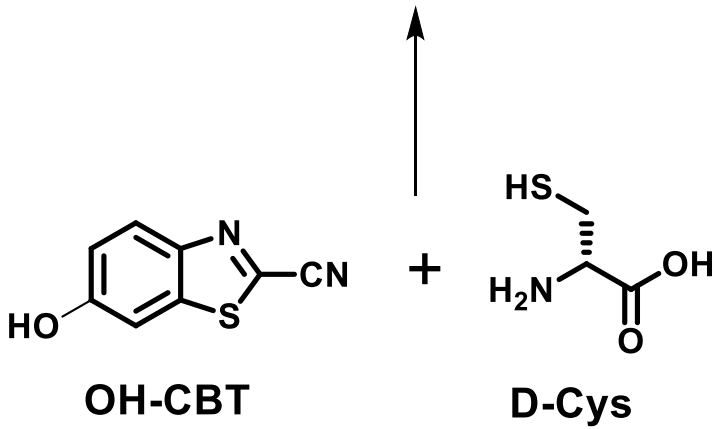


Godinat et. al, ACS Chem. Biol., **2013**, *8* (5), 987–999

Split Luciferin Approach; Advantages



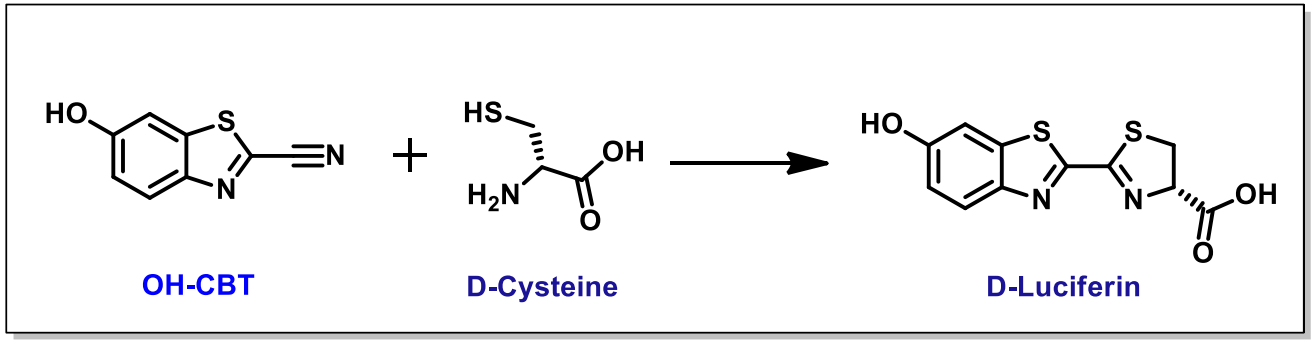
Luciferin

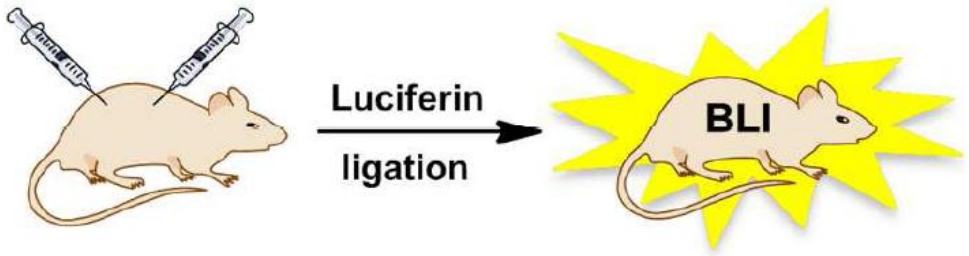


OH-CBT **D-Cys**

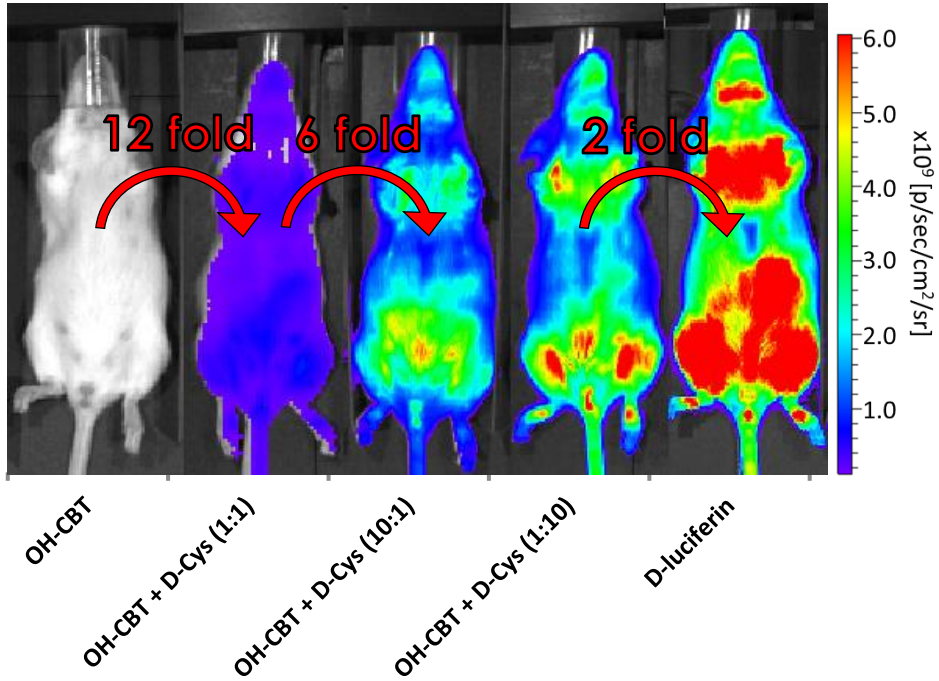
- Higher permeability
- Easier chemistry
- Simultaneous detection
- Better stability
- Lower cost

Does this reaction work in vivo ?





IP injection
FVB-luc+ mice
Dose : 0.268 mmol/kg

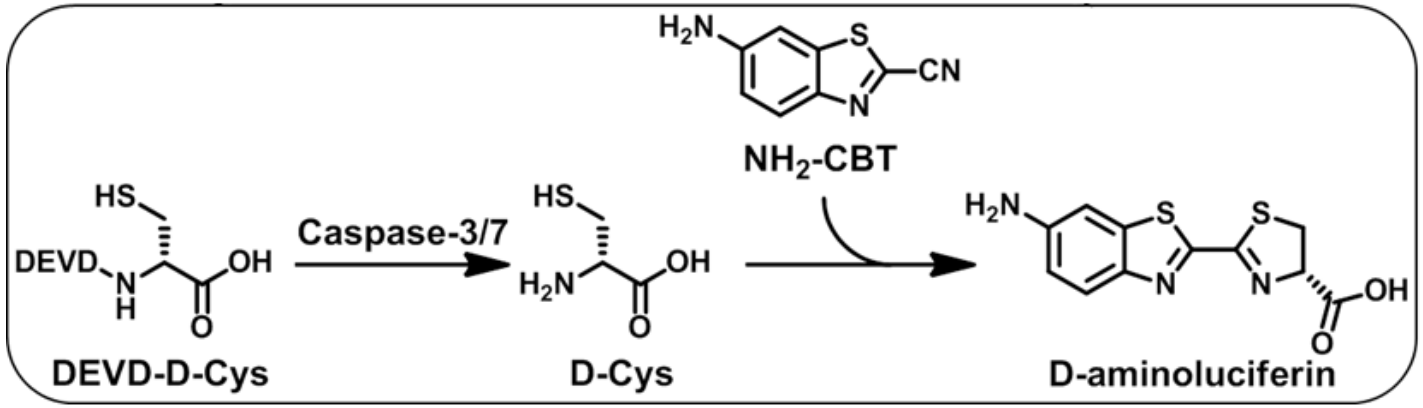


15 min post-injection

In collaboration with Doug Hanahan

Godinat, A. et al, ACS Chem. Biol., **2013**, *8* (5), pp 987–999

Caspase-3 activity imaging in vitro



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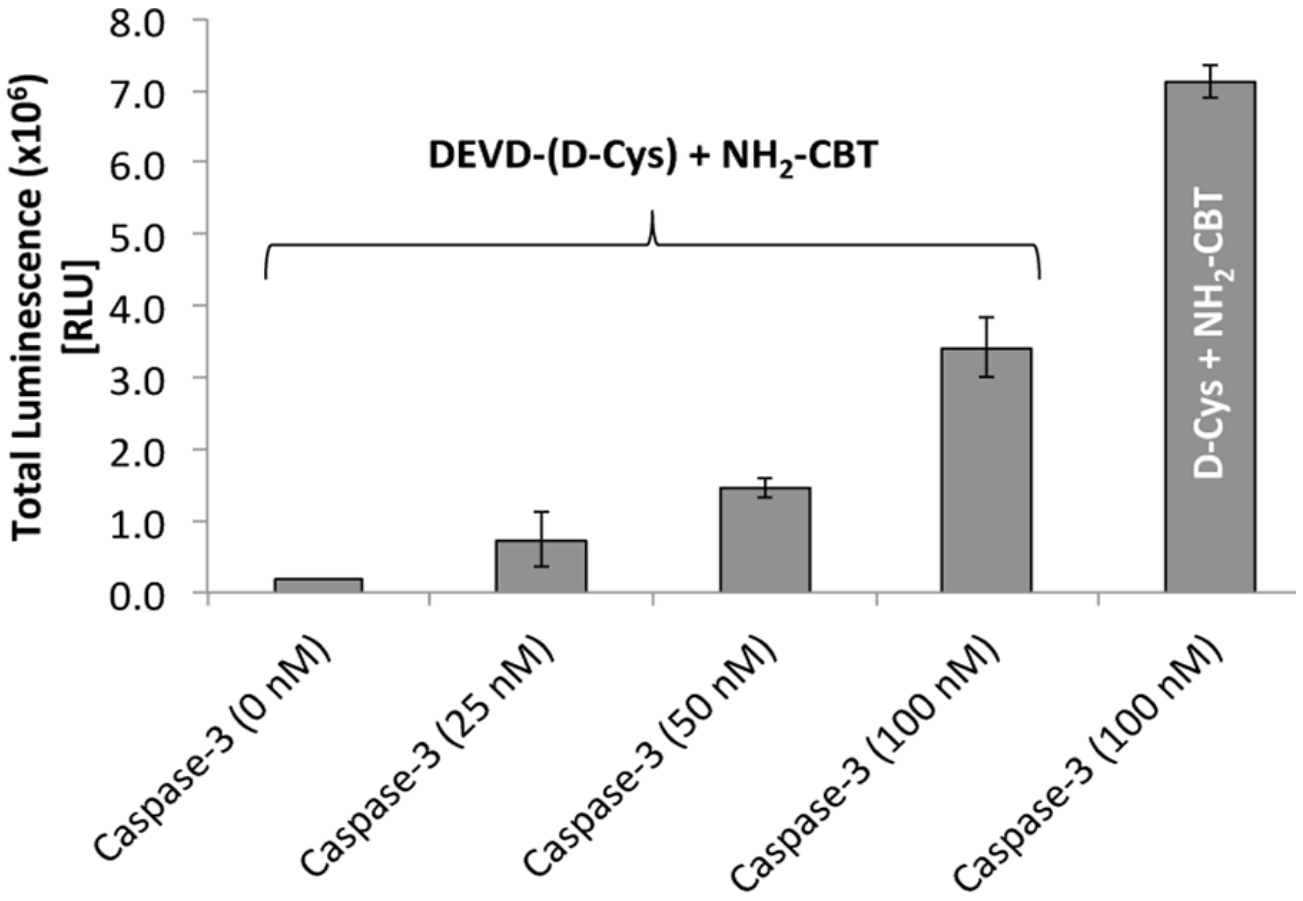
Caspase-3 activity imaging in vitro



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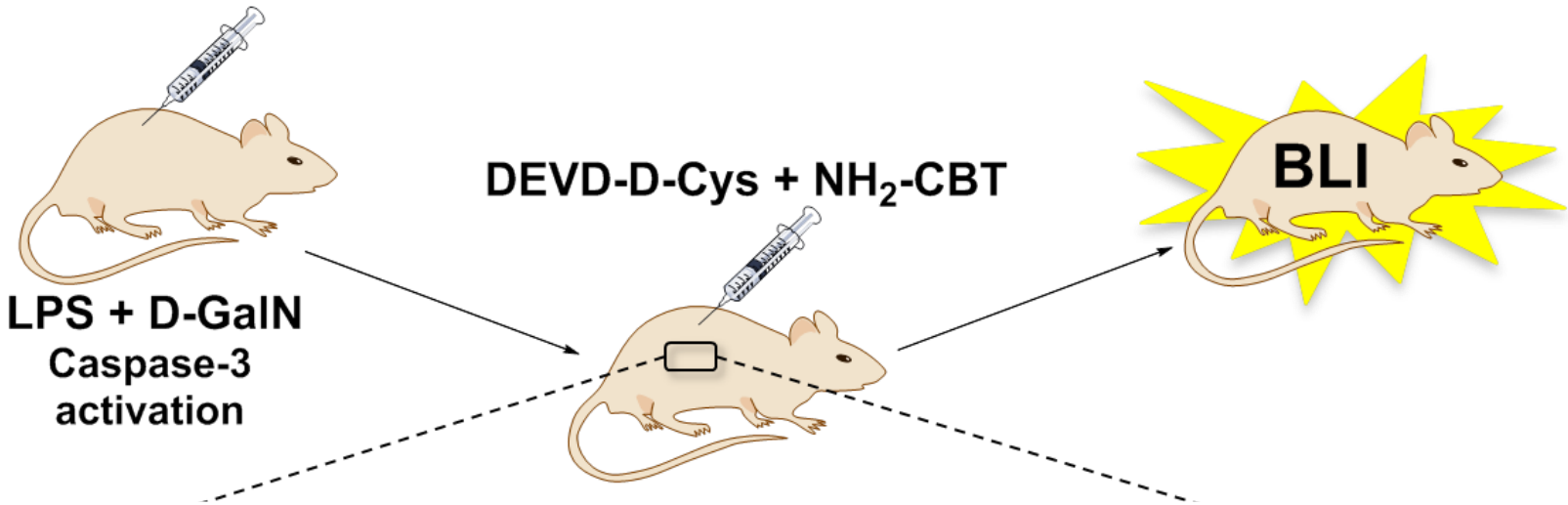
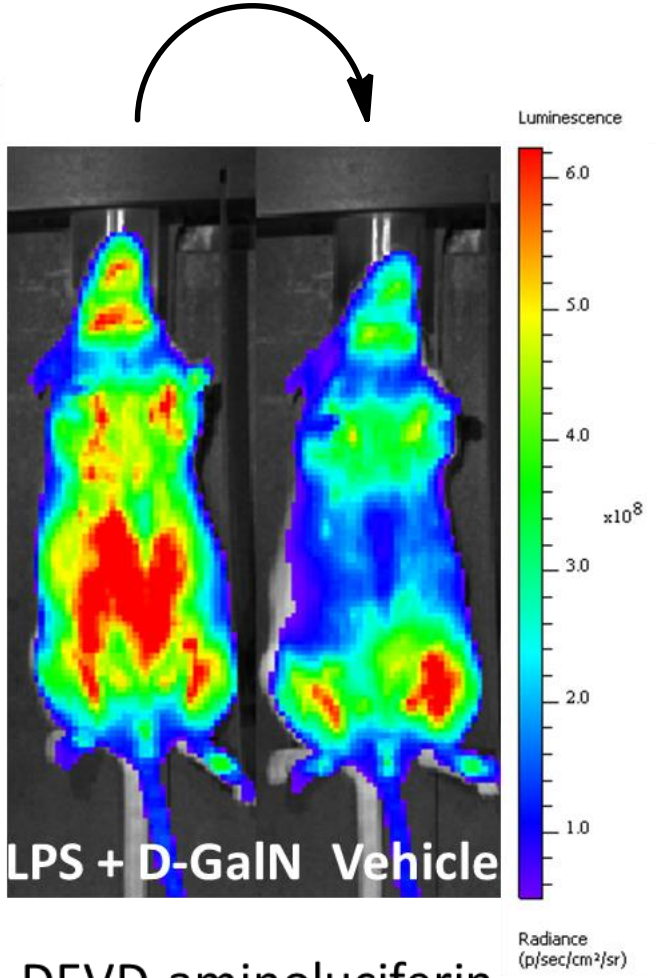
Caspase-3 activity imaging in FVB+luc Mice



Caspase-3 activity imaging in FVB+luc Mice

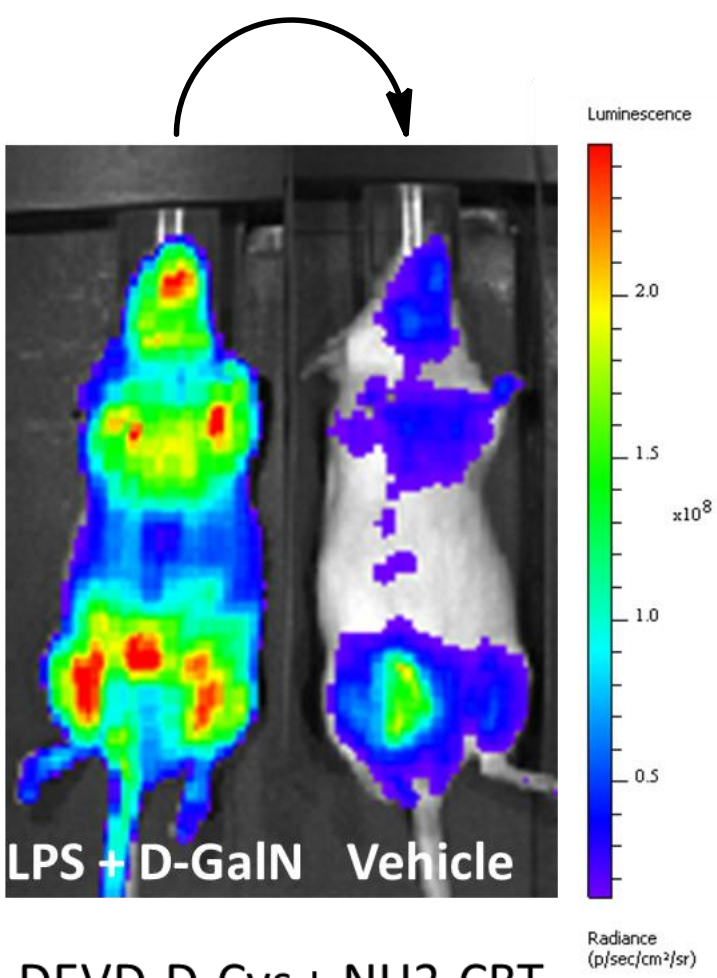
3 fold



DEVD-aminoluciferin

$60/mouse/inj.

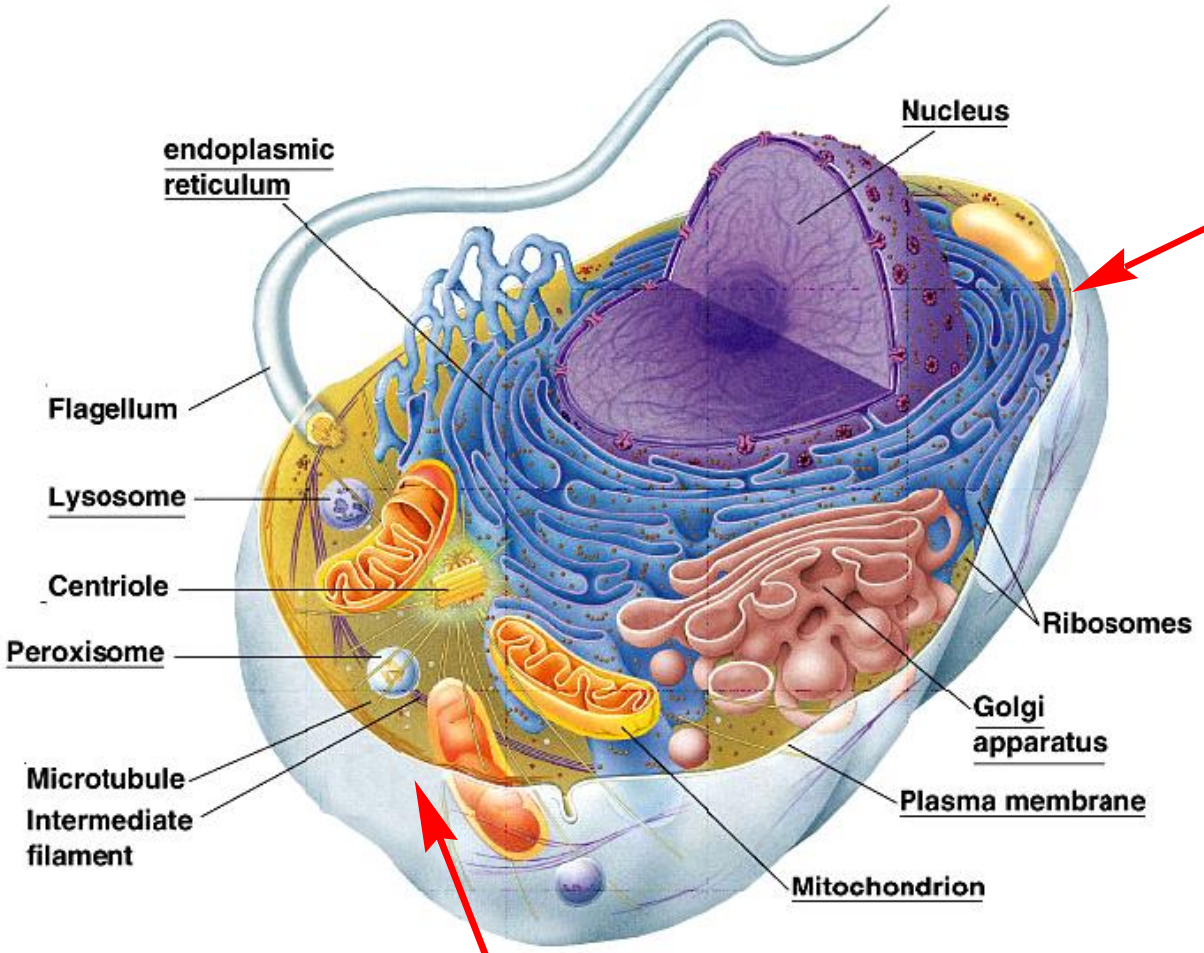
6 fold



DEVD-D-Cys + NH2-CBT

$2/mouse/inj.

Conclusions

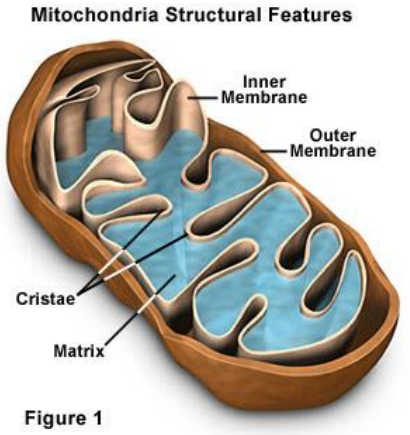


Fatty Acid Transport

Protein functions

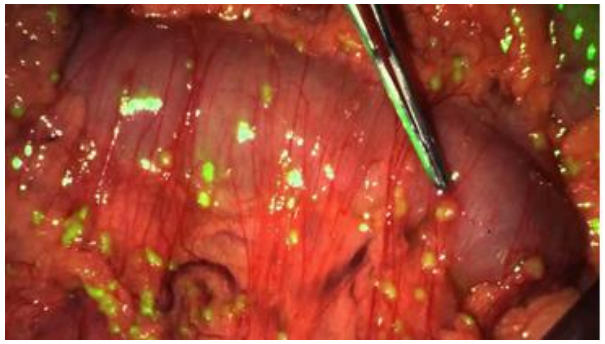
Protease activity

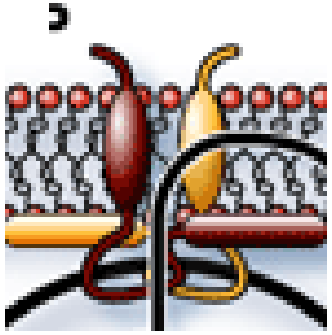
Development of new probes



Mitochondria membrane potential

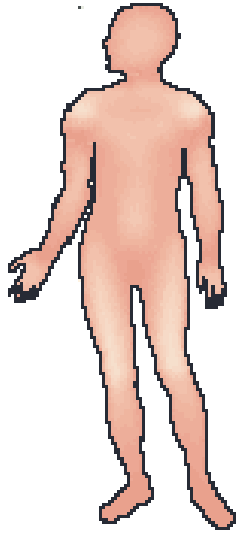
Image guided surgery





Activity of Glucose Transporters

Tumor Imaging



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- Prof. Matthew Bogyo and Laura E. Sanman
  (Stanford University, California)

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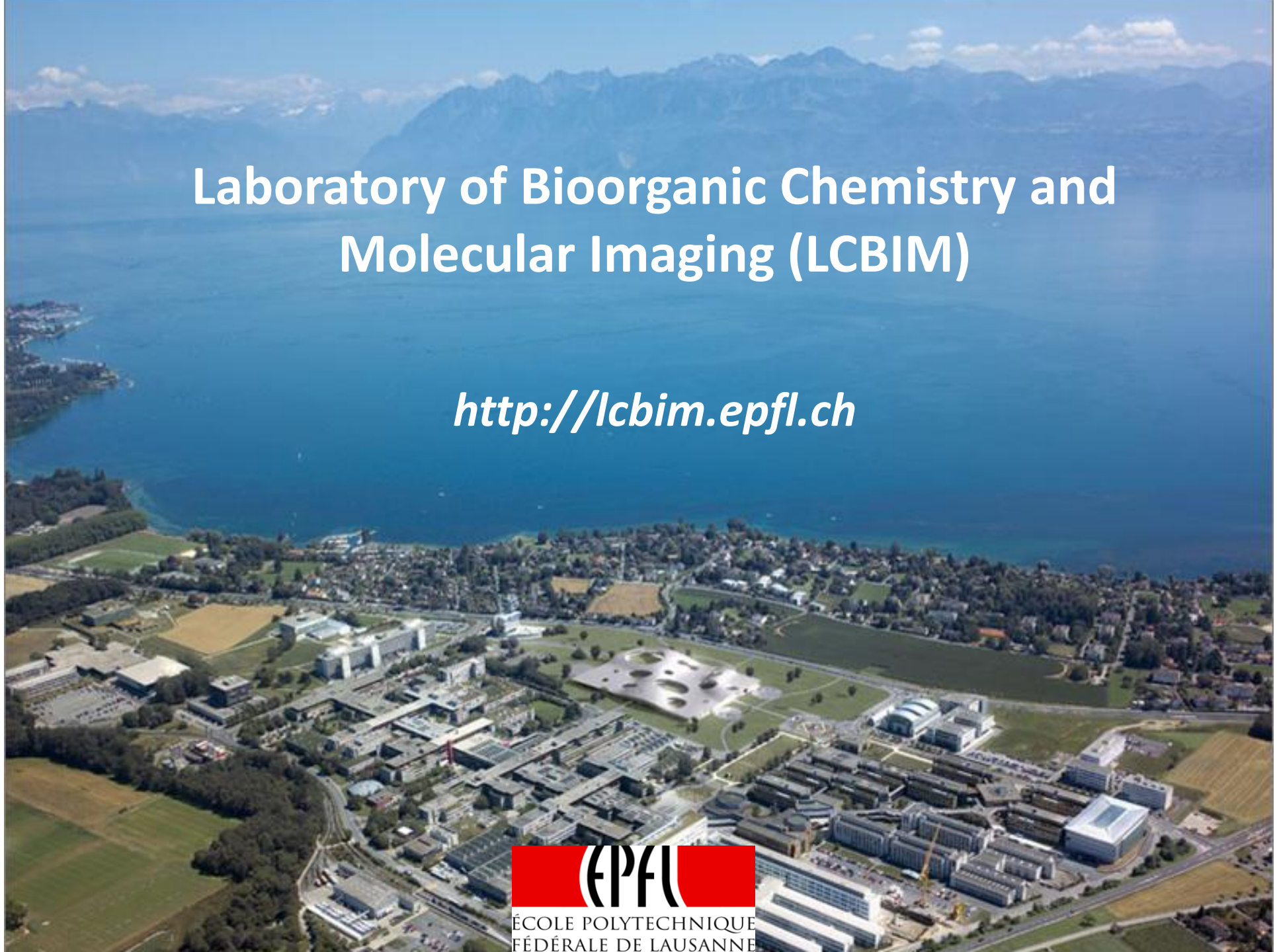
http://lcbim.epfl.ch



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