### Imaging Frontier Center Period: from April 1, 2015 to March 31, 2020

#### **Members**

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	& Tech.
Visiting	Hideo YOKOTA, Visiting Prof., RIKEN Center for Advanced Photonics
Researchers :	Naoko OHTANI, Prof., Grad. School of Medicine, Osaka City Univ.
	Takumi HIGAKI, Assoc. Prof., Int'l Res. Org. for Adv. Sci. & Tech., Kumamoto Univ.
	Keisuke Isobe, Visiting Researcher, RIKEN Center for Advanced Photonics
	Masaya ISHIKAWA, Visiting Researcher, Univ. of Tokyo

#### Purpose

· To research and develop leading-edge imaging technologies for biology and life sciences

• To expand the horizon of imaging research by promoting collaborative research and by exchanging technologies and information among young researchers

#### **Research Subjects**

- Development of imaging technologies with which we can visualize the reactions, temperature and hardness of the microstructures of living bodies as multidimensional information
  - Laser-induced surface deformation for visualizing the dynamic properties of a cell
  - Fluorescence nano-thermometry for thermal imaging
  - Observation of single molecule in order to explore the mechanism of enzyme action

• Development of new technologies that exclude obstacles in the observation pathway to realize fluorescence imaging at deep observation depths

- Visualizing blood vessels, living tissue, and organs in real time to diagnose diseases
- Autofluorescence-free imaging of plant tissues

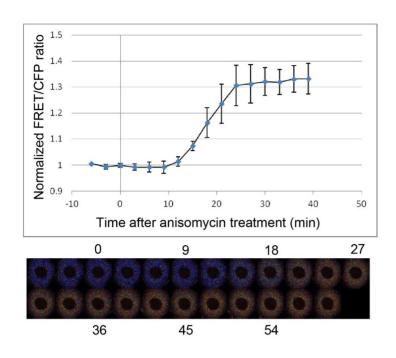
• Development of the application of imaging technologies for research into nervous systems, immune systems, living animal bodies, plants, and farm products.

#### **Present Status and Future Prospects**

 $\cdot$  We have already established a cooperative system inside and outside the TUS through past activities.

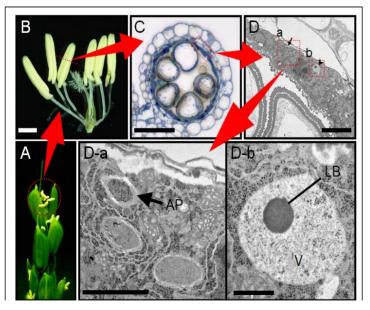
• We are currently planning to form a center of excellence for research through developing frontier imaging technologies and applications. Furthermore, we will extend the use of imaging technologies by widely circulating information about our activities to researchers inside and outside the University .

# Development of green-red FRET sensors for two-photon excitation microscopy



For in vivo FRET imaging, two factors are main obstacles: cross-excitation and a high level of scattering. Here we tried to use a green-red fluorescence protein pair to overcome these difficulties. We have used a green-red sensor for visualizing JNK activity due to its large dynamic range. The left panel shows a representative timelapse images and the increase in FRET/CFP ratio, which were obtained using two-photon excitation microscope. We think that this sensor provides a good proof of concept.

## Imaging of autophagy in the tapetum cells of rice anthers and discovery of its physiological significance in pollen maturation and crop production



Programmed cell death in the tapetum cells, the most inner laver of anthers critical roles plays in pollen maturation. Bv applying various imaging techniques. we have discovered that autophagy is induced at a specific stage and plays a crucial role in pollen maturation and crop production in rice (Kurusu *et al.* 2016, 2017)