Imaging developmental switches during growth and protein interactions in response to stress using FRET-FLIM and STED.

Vinicius Lube, Fatema Aljedaani, Yang Zhang, Shyam Gundu and Ikram Blilou

King Abdullah University of Science and Technology (KAUST), Thuwal, 23955-6900, Saudi Arabia
Email: Ikram.blilou@kaust.edu.sa

Keywords:
Live imaging, in vivo FRET-FLIM, protein complexes, nuclear bodies, STED, stress,

Abstract:
Developmental switches are key to growth and morphogenesis in complex organisms. Being subjected to continuous environmental changes, plants have developed a remarkable developmental plasticity to cope with unpredictable environmental stresses. Here we use a wide spectrum of live imaging technologies to monitor plant growth and organogenesis and profile changes in surface morphology and roughness during and after stress recovery (Figure 1A-B).

We also use FRET-FLIM technology to show spatial protein complex distribution in living tissues during growth and response pathogens. We demonstrate that the newly identified fluorophores improved expression levels, brightness and imaging resolution of nuclear factors. Our findings highlight the importance of fluorescence lifetime imaging and superresolution microscopy to show interactions during growth and defense and to monitor changes in localization and subcellular distribution of co-localized proteins in a subset of living cells. Co-expression studies using STED and interaction analysis using FRET-FLIM showed that developmental regulators and defense proteins colocalize and interact in nuclear compartments (Figure 1C).

Figure 1: Visualizing growth and cellular dynamics in living cells. A, 3D image showing root primordia. B, 3D root profilometry showing a root under normal conditions (left) drought stress (middle) and recovery (right). C, FRET-FLIM showing reduction of fluorescence lifetime in nuclear bodies of a developmental regulator and a defense protein.