Trends in Plant Science | Technology of the Month

Non-invasive live ROS imaging of whole plants grown in soil

Yosef Fichman[1], Ron Mittler[1,2,*]

[1]The Division of Plant Sciences and Interdisciplinary Plant Group, College of Agriculture, Food and Natural Resources, Christopher S. Bond Life Sciences Center University of Missouri, 1201 Rollins Street, Columbia, MO 65201, USA
[2]Department of Surgery, University of Missouri School of Medicine, 1 Hospital Drive, Columbia, MO 65212, USA

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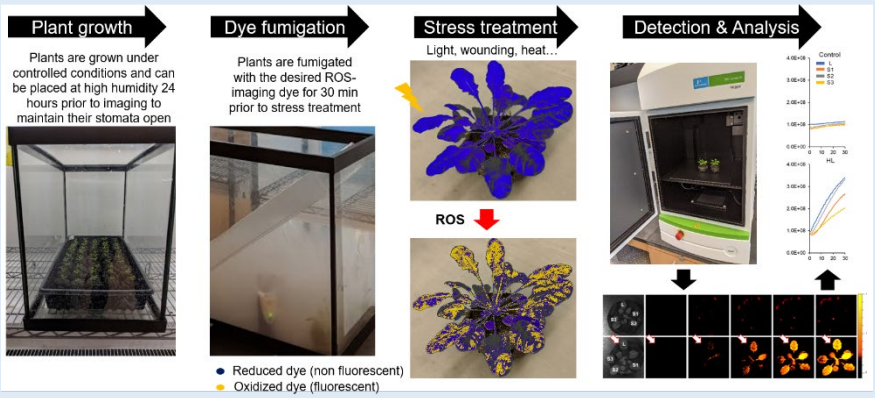
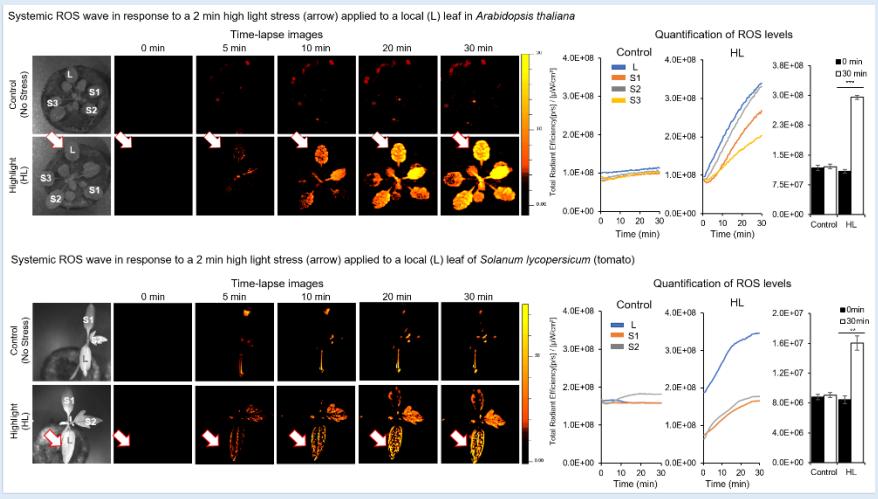


Reactive oxygen species (ROS) play a pivotal role in the biology of all aerobic organisms. Imaging of ROS in plants is critical to our understanding of plant responses to many different environmental and developmental cues. We developed a non-invasive technique to detect ROS in different plant species without the need to transform plants with a ROS reporter or sensor. Different dyes are fumigated into plants prior to a stress treatment and their fluorescence is imaged using an IVIS platform, initially developed to image different florescent proteins in mice.



Using this method, we can detect systemic ROS accumulation in response to different stresses applied a local leaf (*e.g.*, following a local treatment of excess light stress we measured the whole plant increase in dye oxidation over time; Student t-test, SE, N=12, ***P < 0.005, **P < 0.01). The simplicity of the method allows it to be used with different crop plants, as well as to screen different *Arabidopsis thaliana* mutants.

Advantages:
The new method is fast and does not require plant transformation. It could be used for the screening of entire mutant libraries or different plants that are not easily transformed.

Soil-grown plants can be easily imaged allowing for the study of whole-plant responses at different developmental stages.

Different dyes, specific for different types of ROS, could be used with this method to dissect different forms of ROS signalling.

Systemic responses of whole plants or whole plants that are the outcome of different grafting combinations could be studied allowing the advanced study of systemic signalling.

Responses to different biotic or abiotic challenges (as well as mechanical injury), and their combinations could be easily studied

Challenges:
The method is based on the use of the IVIS Lumina S5, or a similar high resolution detection platform, which limits it availability.

The IVIS chamber is limited in size, restricting number of and/or size of plants that can be imaged. Future developments and platform adjustments may solve this issue.

The oxidation of many of the ROS dyes, currently available, is irreversible allowing for the measurements of ROS accumulation processes only. Development of reversible ROS dyes would address this problem.

Although the dyes used are highly sensitive, they have a threshold of signal to noise ratio setting a ROS detection limit for the method. Newer dyes with a higher detection limit would address this issue.

*Correspondence:
mittlerr@missouri.edu (R. Mittler)

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