Monitoring plant leaf response under ozone stress based on OCT biospeckle signal.

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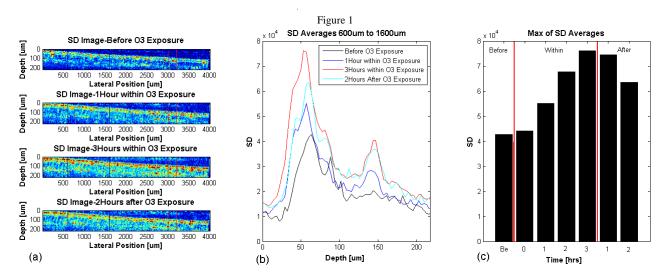
Introduction

To evaluate environmental stress on plants it is important to monitor short-term and long-term influence on plant physiology and plant vitality. Recent studies show that ozone (O_3) stress can cause physiological changes, as well as changes in growth patterns in plants¹. Conventional techniques for studying these changes in plants require destruction and subsequent analysis, thus plant's responses to external stress are inferred indirectly. In this study, we present an optical coherence tomography (OCT) for studying O_3 stress in plants. OCT is an interferometric method that can provide non-contact two, or three dimensional in-vivo tomographic images of the internal tissue structure with very high resolution, in the range of few micrometers². In the experiment, Allium tuberosum commonly known in Japan as Nira was exposed to O_3 and the effect was observed by OCT. In addition to a normal stractual observation by OCT, a biospeckle signal (a dynamic signal due to the movement of organelles inside the leaf) was utilized as a functional visualization. The Standard deviation (SD) of OCT biospeckle signal was found to reflect the influence of ozone well.

Experiments and discussion

The optical system applied in this experiment is a fiber based Michelson interferometer with a SLD at central wavelength 836.1 nm and axial resolution (depth resolution) was estimated to 6µm. OCT imaging data were collected subsequently before O_3 exposure, 1 hour, 2 hours, 3 hours after started O_3 exposure and, 1 hour, 2 hours after stopping O_3 exposure. Both the temperature and the humidity were kept constant during a single session of experiment with respective variations, around $\pm 1\%$ and $\pm 2\%$.

Fig. 1 (a) shows two dimensional standard deviation images of the selected leaf (10Hz scanning frequency in lateral direction) before, 1 hour after started O_3 exposure, 3 hours after started O_3 exposure, and 2 hours after stoping O_3 exposure. SD image represents the standard deviation (SD) of OCT temporal signal (biospeckle signal) obtained at each point in the cross-sectional image. Fig. 1 (b) shows standard deviation (SD) of OCT biospeckle signal for the region 600 to 1600µm and Fig. 1 (c) shows their maximum values attained at palisade tissue (60µm in depth) as a function of time.



We examined the vitality of the plant by looking at temporal development of the signal at 600-1600µm lateral region. As it can be seen from the SD images that, fluctuation become strong with O_3 exposure while reducing after the exposure for the surface and up to 200µm depth at 600-1600µm lateral region. This result indicates that Allium tuberosum plant showed biological responses against O₃ exposure and OCT biospeckle imaging method is capable of monitoring those changes. We presented an optical method that can be used for *in-vivo* studies of plants under environmental stresses. As a preliminary experiment we presented results from Allium tuberosum under O_3 stress. We could observed the response of the plant which differs before, within and after O_3 exposure.

References

1. Rathnayake P. et.al, Journal of Forest Research, 12, 393-402, 2007

2. Fujimoto J .et. al, Optical Coherence Tomography: Technology and Applications, 2008