Abstract
Vis/near infrared imaging analysis for monitoring water condition in plant leaves

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Introduction

Water uptake is one of the most important factors for plant growth. Lack of the water or high osmotic pressure within the soil disturbs the water uptake, and induces a decrease in moisture content of the leaf. Measuring the water content of leaves is a convenient method of evaluating water stress, but it is normally a destructive measurement. Near infrared (NIR) light at the wavelength of 1450 nm is strongly absorbed by water, which is assigned to the first overtone of O-H stretching. Hence the water content might be predicted non-destructively by using the NIR method. Another parameter of evaluating environmental stress is chlorophyll fluorescence induction (CFI). It is a sensitive indicator of photosynthesis, especially related to the electron transport system. In this study these two kinds of image analysis were examined to evaluate the water status in plant leaves.

Materials and methods

Prediction of water content in leaf using NIR imaging

Sixteen leaves were taken from a *Ligustrum japonicum* tree. These leaves were naturally dried at room temperature for four days. During the drying process, NIR images were taken, and weights measured four times. NIR images at $\lambda = 1450$nm were captured by a Visible-NIR vidicon camera (C2741-03, Hamamatsu Co., Japan), using a narrow band-pass filter with central wavelength of 1450 nm, and bandwidth half maximum height of 15 nm. The oven-dried weight was taken immediately after each series of NIR measurements. Finally the NIR data from the reference region were subtracted from those of the sample region to compensate for drift. Histograms were developed for the average cumulative differences in luminance (CDL) between the sample region and reference regions. The cumulative frequencies were regarded as explanatory variables, and partial least squares (PLS) regression was performed to predict water content of the leaves. One half of the data set was employed for calibration, and the other half was used as a prediction set.

Time-resolved PCA imaging for CFI

Potted *Epipremnum aureum* were used for this experiment. The pots were put in a thermostatically-controlled chamber for 24 hours. The temperature and humidity were maintained at 25°C and 60%, respectively. After one hour at a dark condition, the plants were sequentially irradiated with blue light (470 nm) and red light (640 nm) to induce chlorophyll fluorescence. Time resolved CFI images at $\lambda = 720$ nm were captured by a CCD video camera module (XC-ST50, SONY, Japan) with a narrow band-path filter. Time resolved images of CFI were captured at 3 fps (frames per second) for 20 seconds. After acquisition of the first image, some leaf stems were cut, to stop the water supply. Image acquisition was performed at no hours, two hours, four hours, six hours and eight hours after cutting. Then the time- resolved profile of fluorescence intensity of cut leaves and water-supplied leaves were developed. PCA was performed, using luminance brightness at each frame as explanatory variables.

Results and discussion

Prediction of water content in leaf using NIR imaging

The cumulative histogram increased in terms of the *CDL* as water content decreased.

Table 1 shows the PLS predicted results calculated from the cumulative histograms for *CDL*. PLS analysis using cumulative histograms for *CDL* could predict the water content in leaves with satisfactory accuracy.

Time-resolved PCA imaging for CFI

The first notable peak on the time resolved profiles of *CFI* decreased as water stress increased. On the other hand, fluorescence intensity after 10 seconds increased (Figure.1). A score plot of PC1 versus PC2 could separate the water stressed leaves from healthy leaves (not shown here).

Table 1. PLS regression result calculated from cumulative histograms for *CDL*.

N	32
PC	2
R^2	0.90
RMSEP (%)	28.8
RPD	3.27

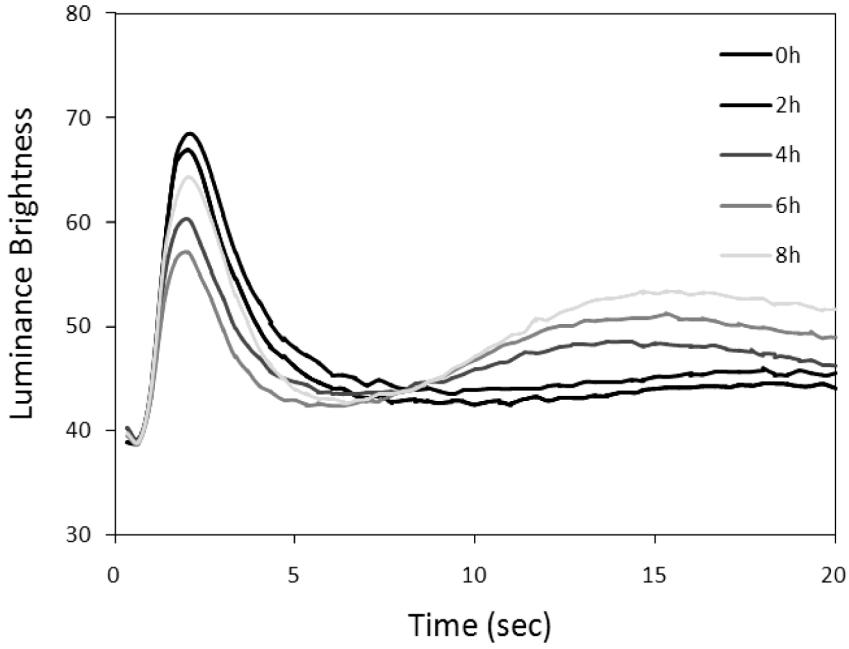

Figure 1. Time profiles of fluorescence intensity in leaves.