
Ground-based Spectral Measurements of Chlorophyll

Fluorescence from Vegetation Canopies

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Abstract

Detection of vegetation fluorescence is important for understanding physiology of vegetation leaves in relation to photosynthetic activities. We have developed a stand-off system that enables the observation of vegetation fluorescence under both laboratory and field conditions. The intrinsic difficulty in monitoring weak fluorescence signal is the separation between the fluorescence signal around 740 nm and the large reflection of vegetation leaves in the near infrared (NIR) spectral region. In the laboratory measurement, it has been proven that a LED-based light source coupled with cold- or hot-mirrors can provide illumination with only visible or NIR part of the spectrum. The “pure” visible illumination, which is mostly free from the NIR radiation, can lead to the observation of fluorescence signals without hindrance from large NIR reflection. The NIR illumination, on the other hand, enables the evaluation of pure reflectance in NIR free from the fluorescence contribution. The combination of such pieces of spectral information makes it possible to implement the detailed analysis of solar radiation induced fluorescence signals. For field applications, a stand-off measurement system based on an astronomical telescope has been developed to carry out the spectral measurement using a CCD spectrometer, together with the two-dimensional measurement of the fluorescence intensity distribution by means of a cooled CCD camera. A narrow-band optical filter centered at 760 nm, the wavelength of the oxygen A-band, is employed with the CCD camera to exploit the “solar blind” wavelength for the fluorescence measurement under daylight conditions. We describe the application of the system to a soya-bean field in Kyoto University.

Keywords

Vegetation fluorescence; Ground-based remote sensing; Canopy measurement;

1. Introduction

Chlorophyll fluorescence emitted from vegetation under insolation yields valuable information on photosynthetic activities. It has been reported^{1,2)} that high-resolution FITR spectrometers onboard satellites are capable of detecting fluorescence signals from surface

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vegetation. For ground-based observations, instrumentations such as the use of UAVs³⁾ and a crane tower above fields⁴⁾ have been proposed and tested.

The present paper reports the results of our recent activity toward the realization of a system that enables remote sensing of vegetation fluorescence on the canopy level. The system is composed of an astronomical telescope, a compact CCD spectrometer, a cooled CCD camera, and narrow-band filters^{5,6}. In order to carry out quantitative analysis, it is indispensable to validate the signal against both fluorescence and reflectance signals in the near infrared (NIR) region taken inside the laboratory, since the separation of these two contributions is essential. When an LED light source is employed in such a laboratory experiment, it is shown that the use of a cold mirror (good rejection of NIR and transmission of visible light) or a hot mirror (good rejection of visible light) can lead to ideal illumination conditions for separate observations of weak fluorescence signals and large signals arising from infrared reflectance. As an example of our recent field studies, here we describe the results of solar radiation induced fluorescence (SIF) conducted at a soybean field in Kyoto University with stand-off distances of 15-30 m.

2. Laboratory observation of fluorescence spectra

The light from an LED light source (35 W) was illuminated on a soybean (*Glycine max*) canopy prepared in a dark laboratory. Figure 1 shows the LED spectrum, which exhibits peaks around 450 and 550 nm, with relatively weak intensity at wavelengths longer than 700 nm (broken line). For further reducing the NIR intensity, the combination of a cyan filter and a hot mirror (transmitted light) was used to cut wavelengths longer than 620 nm (thin curve in Fig. 1). The resulting LED-induced fluorescence signal of the vegetation canopy exhibits a major peak at around 690 nm with a smaller peak at 690 nm (solid curve).

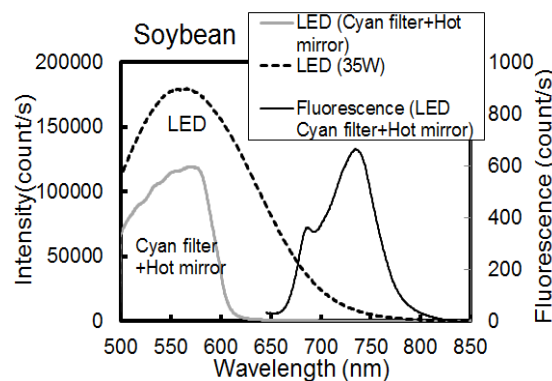


Fig. 1 Spectra of original and modified (cyan filter + hot mirror) LED light source shown with LED-induced fluorescence signal.

3. Stand-off measurement of SIF

The field measurement of soybean SIF was carried out at the experimental field of Kyoto University during September 1 -3, 2016. The cultivated Soybean types are Toyoyutaka and some other varieties. The observation under daylight conditions were carried out with an optical system developed in our group^{5,6}. The main feature of the system is the capability of sharing nearly the same field-of-view (FOV) of a telescope with a compact spectrometer and a cooled CCD camera in a subsequent manner (Fig. 2) The diameter of the telescope is 200 mm, with an FOV coverage of 0.47 deg (full angle). The spectrometer is a cooled CCD spectrometer (Ocean Optics, QE65Pro) covering the wavelength range of 500-870 nm with a sampling interval of 0.1 nm. Typical value of the signal-to-noise ratio is as high as 1000:1. The spatial distribution of fluorescence intensity is detected with a cooled CCD camera (Bitran, BU51LIR) equipped with an optical filter centered at 760.68 nm with narrow-band transmission of 1 nm (FWHM) (Andover, hereafter F760).

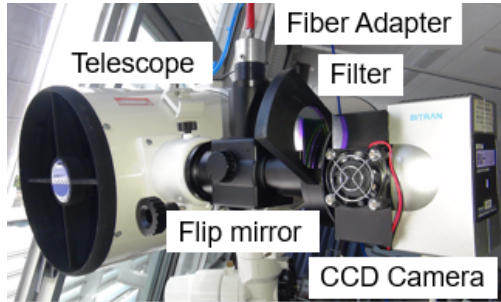


Fig. 2 Instruments used for the field observation of SIF

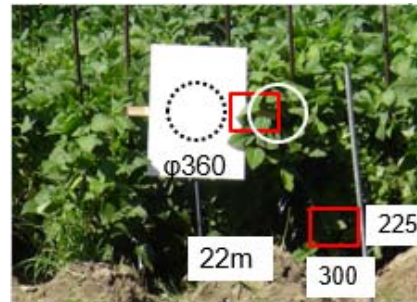


Fig. 3 Locations of the canopy and reference (whiteboard) observation.

For obtaining reference spectrum/image, the spectrum of solar radiation reflected from a home-made whiteboard (WB) of $\sim 0.45 \times 0.6 \text{ m}^2$ was measured with the telescope system, for both the spectrometer and camera image measurement modes. Figure 3 shows the scheme of the reference measurement. In the case of the spectral measurement, the portion of the WB indicated by a dotted circle was employed and the resulting spectrum was compared with the vegetation signal taken by observing the position of the soybean leaves encircled with the white line. In the case of the image measurement, the rectangular area of $\sim 0.30 \times 0.23 \text{ m}^2$ (marked with red line) was observed, the left part of which is covered by the WB.

Figure 4a shows the resulting spectrum from both soybean leaves (thin line) and the WB reference (solid line). In the spectral region of 825 - 850 nm, the effects of SIF and water vapor absorption are insignificant. Thus, the reflectance of target leaves relative to the reference WB is estimated from this region, as indicated with a dotted curve. Here the slight decline of WB reflectance toward the shorter wavelength was taken into account. Then, the difference between the soybean curve (thin line) and the scaled reference curve (dotted line) indicates the SIF signal, as illustrated in Fig. 4b. For the following discussion on the SIF intensity, we use the spectral intensity at 775 nm, which is mostly free from the effects of atmospheric absorption (i.e., O_2 and water vapor) and chlorophyll absorption.

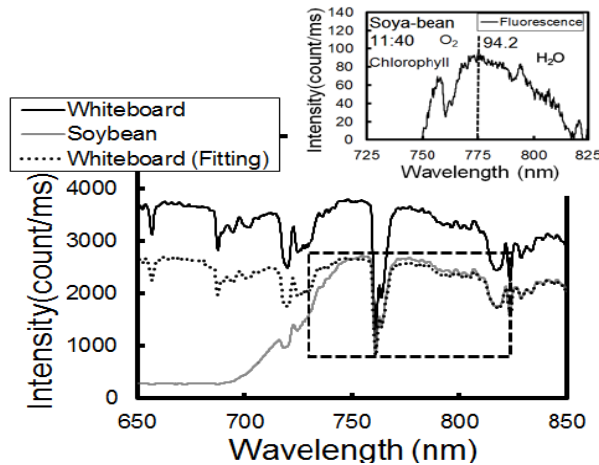


Fig. 4 Proposed Method of spectral fitting. In (a), the WB spectrum (solid line) is scaled to fit the vegetation spectrum (thin curve) using the wavelength range of 825 - 850 nm. Then, the resulting curve is subtracted from the canopy signal to obtain the SIF profile, as shown in the inset.

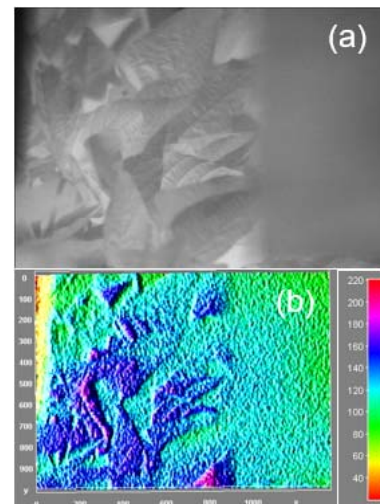


Fig. 5 (a) Soyabean field imagery and (b) fluorescence intensity calculated by subtracting the reference spectra after the scaling.

Figure 5 shows the spatial distribution of SIF. In this image, the right side shows the reflectance of the WB, while the left side the reflectance from the soybean canopy. The camera image was recorded by applying the narrow bandpass F760 filter. We assume the linearity between the pixel digital numbers (DNs) and radiation intensity, and the same scaling factor obtained in the case of Fig. 4a is applied to this case as well. Thus, the difference between the canopy DN and scaled reference DN gives the SIF intensity distribution as indicated in Fig. 5b.

The temporal change of the SIF intensity from both the spectral and camera measurements was compared with that of photosynthetically active radiation (PAR) recorded using a PAR meter (BMS 3415FXSE) (Figure not shown). Good agreements were seen among these three independent datasets, suggesting the direct relation between the PAR and fluorescence intensities during the observation time.

Figure 6a shows the reflectance image taken with the CCD camera attached with an interference filter having the central wavelength 550 nm and width 10 nm. The stand-off distance was 20 m, and the acquisition time duration was 30 ms. In this visible image, it is seen that generally strong reflection of solar radiation is observed for leaves that meet the specular reflection condition. Figure 6b, on the other hand, shows the SIF image taken by applying the F760 filter with the acquisition time of 50 ms. The color scale is assigned for the data expressed in units of $\text{count ms}^{-1} \text{ pixel}^{-1}$.

In the experimental field of Kyoto University, different varieties of soybean are planted column by column. Figure 7b, the red squares indicate the position of columns. As seen from Fig. 7, good agreement was seen for the SIF intensities determined from the spectral measurement and image analysis.

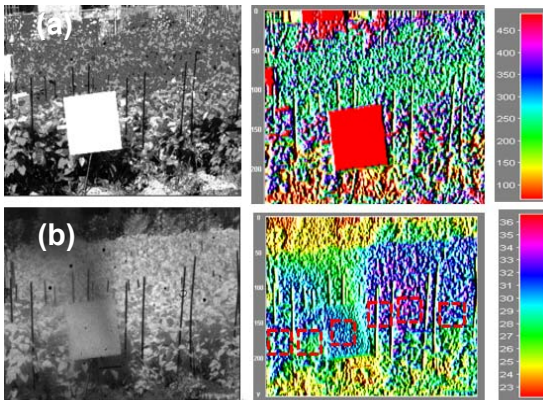


Fig. 6 (a) Reflection image observed at 550 nm and (b) fluorescence image observed at 760 nm.

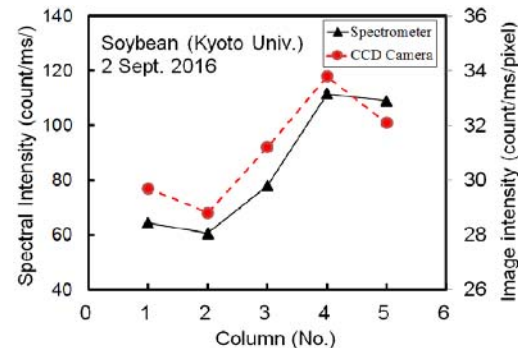


Fig. 7 Difference in fluorescence intensity observed for different varieties of soybean (14:10-14:30 on September 2, 2016)

In conclusion, we have described the SIF measurement from vegetation canopy under both laboratory and field conditions. The use of LED light source coupled with appropriate filter setup made it possible to separate the weak fluorescence intensity from much larger near-infrared reflectance of vegetation. In the field measurement, the scaling of the whiteboard reference worked well for deriving the fluorescence signals even under daylight conditions.

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