

## THE EFFECTIVE OF DIFFERENT EXCITATION WAVELENGTHS ON THE IDENTIFICATION OF PLANT SPECIES BASED ON FLUORESCENCE LIDAR

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### ABSTRACT:

Laser-induced fluorescence (LIF) served as an active technology has been widely used in many field, and it is closely related to excitation wavelength (EW). The objective of this investigation is to discuss the performance of different EWs of LIF LiDAR in identifying plant species. In this study, the 355, 460 and 556 nm lasers were utilized to excite the leaf fluorescence and the fluorescence spectra were measured by using the LIF LiDAR system built in the laboratory. Subsequently, the principal component analysis (PCA) with the help of support vector machine (SVM) was utilized to analyse fluorescence spectra. For the three EWs, the overall identification rates of the six plant species were 80%, 83.3% and 90%. Experimental results demonstrated that 556 nm excitation light source is superior to 355 and 460 nm for the classification of the plant species for the same genus in this study. Thus, an appropriate excitation wavelength should be considered when the LIF LiDAR was utilized in the field of remote sensing based on the LIF technology.

### 1. INTRODUCTION

The diversity of vegetation is very important to the entire ecological system. In recent decades, a large number of technologies (Gong, et al. 2012; Vauhkonen et al. 2013), including passive and active remote sensing, have been proposed to monitor vegetation species. For passive remote sensing, spectral reflectance measurements of vegetation territory can provide the relative area covered by vegetation chlorophyll with respect to the total area being scanned (Du et al. 2014). These data are valuable, particularly when monitoring changes in the local vegetation cover. However, the using of the acquired spectral data disables the accurate classification and identification of plant species or groups. In addition, this technology was also restricted by many other factors, such as weather condition, measurement time etc. (Wu et al. 2009). Hence, researchers hope to be able to develop new techniques that can overcome these deficiencies. Then, LiDAR was proposed in the field of remote sensing (Koukoulas et al. 2005). It was not limited by the weather condition and measurement time, and displayed the advantage of high temporal-spatial resolution and non-destructive. Therefore, it has been widely applied in vegetation monitoring. In addition, Gong et al. (2012) proposed multi-wavelength canopy LiDAR (MWCL) containing both the spatial and spectral information for remote sensing of vegetation. This technology has been successfully utilized to distinguish the coniferous from broad-leaf forest and can be employed to monitor the nutrient stress of crops.

In the past decade years, an active remote sensing technology was proposed by Chappelle et al. (1984), which utilizes ultraviolet (UV) light as the excitation light source to acquire plant fluorescent characteristics. As part of active remote

sensing technology, the feasibility of laser-induced fluorescence (LIF) for detecting the status of vegetation has been tested in aircrafts (Rogers et al. 2012). LIF technology exhibits the advantages of quick response, high sensitivity, and non-destructive property (Apostol et al. 2007). Then, LIF has the potential to become a significant approach for vegetation detection (Günther et al. 1994). At present, LIF is usually employed with an emission spectra measured at UV excitation wavelength (EW) because the fluorescence quantum efficiency of the fluorophore at 355 nm is higher than those at other EWs (Agati 1998). A lot of researches have taken place since the early studies of Chappelle et al., where LIF was mainly used to monitor the status and biomass of vegetation with UV excitation (Ramos and Lagorio 2004). Chappelle et al. (1985) analysed the differences among monocots, dicots and hardwoods based on the characteristics of vegetation fluorescence peaks. The LIF is that the energy of a specific wavelength was absorbed by fluorophore and emitted the light at longer wavelengths. Thus, the LIF technology is closely related to EW. At present, a large number of investigations have been conducted on the correlation between fluorescence spectra and different EWs (Yang et al. 2016). To data, however, few studies have been done on the EWs analysis of LIF LiDAR for the classification of plant species based on the principal component analysis (PCA) combined with support vector machine (SVM).

Therefore, this study is mainly to investigate the effect of EW on the identifying of plant species based on LIF technology. The LIF LiDAR was built in laboratory, and three different excitation light sources (355, 450 and 556 nm lasers) were utilized to induce the leaf fluorescence of plant species. These fluorescence spectra data measured were stored in a

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fluorescence spectral database. PCA was utilized to analyse these fluorescence spectra by extracting the major attributes and reducing the dimensionality of variables. It found that over 90% of the total variance can be explained by using the first three principal components (PCs). Then, SVM algorithm based on these PCs served as input variables was employed to identify these plant species included those of the same family.

## 2. MATERIALS AND METHODS

### 2.1 Samples

In this experiment, the leaves of eight typical plant species (including *Cerasus yedoensis*, *Cerasus dielsiana*, *Cinnamomum kotoense*, *Salix babylonica* Linn., *Ilex chinensis* Sims, *Magnolia denudata* Desr.) were gathered from the subtropical zone of central China at 30°32' N and 114°21' E. The data of sample is 11 July 2015. These samples were sealed in plastic bags, kept in an ice chest, and then immediately transported to the laboratory for LIF spectra measurements.

### 2.2 Experimental system

The schematic of the experimental instrument is shown in Figure 1. The instrument consists of three main parts: the signal acquisition part, the optical receiver assembly, and the excitation light source. The UV excitation light source is 355 nm and is emitted by a frequency-tripled Nd:YAG laser. The 460 and 556 nm lasers were manufactured by Spectra-Physics. The three EWs are relatively easy to be obtained, and represent the UV, blue, and green excitation light. Maksutov-Cassegrain telescope was used to collect the emission fluorescence signals of leaf. Then, a single-mode optical fiber with a diameter of 200 μm was utilized to transmit the fluorescence collected between the telescope and spectrograph. The slit of the spectrograph was set to 0.5 mm. LIF signal variation with wavelength was detected by using an intensified charge-coupled device (ICCD) camera. The fluorescence data was stored in a personal computer. In this study, the spectral range of fluorescence, which was excited by 355 nm laser, was 360-800 nm and with a 0.5 nm sampling interval. The fluorescence excited by 460 and 556 nm ranged from 630 to 790 nm and sampling interval was 0.5 nm (Figure 2).

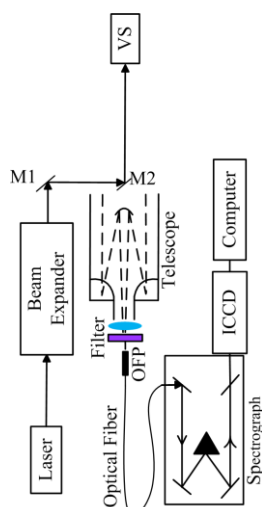


Figure 1. Schematic of LIF Lidar. M1 and M2: completely reflecting mirror, VS: vegetation sample, OFB: optical fiber probe

### 2.3 Data analysis and methods

The fluorescence spectra of leaves were measured by utilizing the LIF instrument (Figure 1), and the data collected were stored in a personal computer. To reduce the systematic error and the effects of laser energy fluctuation, all fluorescence spectra were repeatedly measured nine times and calculated the average. In this study, of which 240 sets of fluorescence spectra data were acquired from each of the three EWs (355, 460 and 556 nm). To reduce the variables of the fluorescence spectra datasets, PCA, a traditional method of multivariate analysis, was utilized to analyse fluorescence spectra (Agarwal et al. 2003)

The PCA is a powerful multivariate statistical data analysis technique. The multiple variables can be converted to a few variables, in which each principal component (PC) is a linear combination of the primitive variables. These PCs extracted the most important information containing in the variables. Thus, this method has been widely used in many research fields. The detail introduce can be found in reference (Yi et al. 2007). Therefore, PCA was employed to extract the feature vectors of fluorescence spectra by analysing major attributes.

SVM was then utilized to analyse the performance of fluorescence spectra excited by different EWs for the classification of plant species. The SVM is a classical supervised learning algorithm with a strong theoretical foundation in statistical theory which can convert low dimensional characteristics to high dimensional characteristics for recognizing some complex targets. The detailed describing can be referenced. The SVM exhibits the special advantages in heterogeneous classes for small samples and high dimensional cases. The kernel function is a critical factor for SVM analysis, and then the radial basis function (RBF) was utilized in this study. It can be presented follows:

$$K(x_i, x_j) = \exp(-\gamma \|x_j - x_i\|^2) \quad (1)$$

Where  $\gamma$  = kernel parameter,

$x_j$  = test data

$x_i$  = training inputs

All new feature variables were then randomly separated into two parts: 75% as the training set to train the SVM model and 25% as the validation set to test the model tentatively. Before analysis, these fluorescence spectra were denoised and smoothed by using a moving-window polynomial fitting and wavelet transform, respectively.

## 3. RESULTS AND DISCUSSION

The 355, 460 and 556 nm excitation light sources were utilized to excite the leaf fluorescence spectra of different plant species. The average normalized fluorescence spectra of all samples of the same species, which was excited by different EWs, are shown in Figure 2. Figure 2 demonstrates that the leaf fluorescence spectra of different plant species were similar at single-excitation wavelength (355, 460 or 556 nm). All of the fluorescence spectra display two main fluorescence peaks bands at 680~690 nm and 730~740 nm. The centre wavelengths of the two fluorescence peaks are 685 and 740 nm, respectively. As concluded by Chappelle et al. (1984, 1985), two fluorescence peaks at 685 and 740 nm are attributed to the centre pigment of Photosystem II (chlorophyll a) and antennae chlorophyll of

Photosystem I (chlorophyll b), respectively. In addition, the fluorescence induced by 355 nm laser exhibited a peak at 440 nm which is caused by ferulic acid and contained more spectra information than that induced by 460 and 556 nm laser. From Figure 2, it can be known that the fluorescence spectra excited by the 460 nm laser exhibited the fluorescence peaks at 685 nm is more intensive than that 740 nm, and the fluorescence characteristics is contrary when 556 nm laser serves as excitation light source. The fluorescence spectra excited by 355 nm laser show inconsistent characteristics of fluorescence peaks for different plant species. These results are similar with the investigations of Apostol et al. (2007) and Agati (1998). Thus, LIF LiDAR can be utilized to distinguish the plant species on the basis of these characteristics of the fluorescence peak.

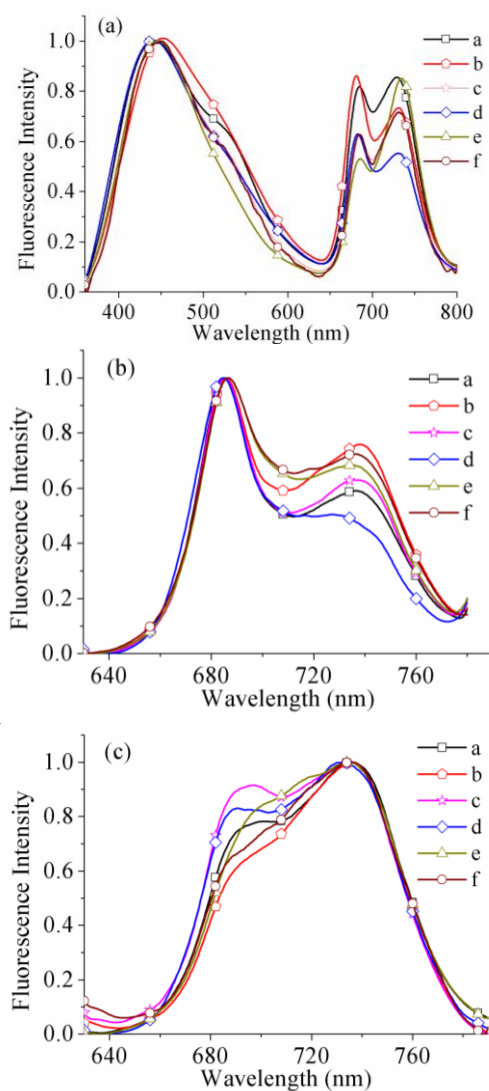


Figure 2. The normalized LIF spectra of the eight plant species were induced by different excitation wavelengths. (a) *C. yedoensis*, (b) *C. kotoense*, (c) *S. babylonica* Linn., (d) *C. dielsiana*, (e) *I. chinensis* Sims, (f) *M. denudata* Desr.

The fluorescence spectra excited by different EWs display different fluorescence spectra features (Figure 2). The reason is that fluorescence will be re-absorbed on its path towards the leaf surface. It has been investigated by Agati (1998) and Ramos and Lagorio (2004) in detail. The previous investigation found that carotenoids and chlorophylls in green leaf have a

broad absorption band in the 400-500 nm spectral regions and the re-absorption process by the chlorophyll pigments in the upper layer leaf cells at emission fluorescence spectra between 730-740 nm is much smaller than that between 680-695 nm. In this study, the 460 nm excitation light will not penetrate very deeply in the leaf and the fluorescence excited by 460 nm laser is mainly generated in the mesophyll cells. Therefore, the re-absorption of fluorescence is weak. The fluorescence excited by 556 nm laser is generated deeper in the leaf tissue than that 460 nm laser. It will result in a longer pathway and the stronger re-absorption. Hence, these fluorescence spectra excited by 460 and 556 nm lasers exhibited different spectral shapes. However, it cannot explain the spectra measured by 355 nm excitation light and still needs to further study in the future.

Then, PCA was used to reconstruct the fluorescent feature vectors and to reduce the dimensionality of the spectra by analysing major attributes. It was found that when the number of PCs exceeded three, the increase of the explained variance with additional PC was reduced to less than 1%. Then, in the three-dimensional coordinate system on the basis of PC1 and PC2, six plant species cannot be distinguished from each other, (Fig. 3).

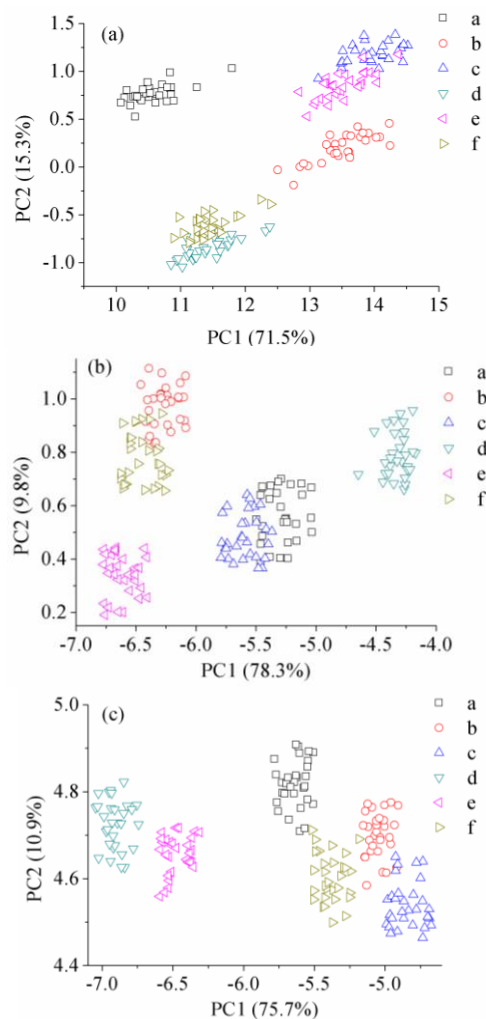


Figure 3. The principle component analysis scores of eight vegetation species under different excitation wavelengths: (a) 355 nm; (b) 460 nm; (c) 556 nm.

In order to comprehensively analyse the ability of fluorescence spectra excited by different EWs for the identification of plant species, the SVM was then employed to distinguish these plant species on the basis of the variables calculated by PCA. 240 sets of experimental data of each EW were randomly separated into two groups: 180 sets designated as the training set for training SVM and the remaining 60 sets employed as the testing set to verify the tentative performance of the model.

#### 4. CONCLUSION

In summary, the fluorescence spectra of eight plant species excited by different EWs (355, 460 and 556 nm) were analyzed and found that different EWs will result in different fluorescence spectra shapes. The probable explanation is that the effect of the re-absorption process of the chlorophyll pigments on the fluorescence spectra. The fluorescence peak at 685 nm is more intensive than that 740 nm when 460 nm laser served as excitation light source, and the fluorescence spectra excited by 556 nm laser display the contrary cases. The fluorescence spectra, which were excited by 355 nm laser, exhibited inconsistent characteristics of fluorescence peaks for different plant species. The specific reason still needs to further study in the future. Then, PCA combined with SVM was employed to analyze the fluorescence spectra excited by different EWs for identifying plant species. When 355, 460 and 556 nm lasers served as excitation light sources, the overall identification rates of the eight plant species were 80%, 83.3% and 90%, respectively. Experimental results demonstrated that the 460 nm EW is superior to 355 nm EW for the classification of the plant species of the same genus, and is inferior to 556 nm EW in this study. Therefore, an appropriate EW should be chosen based on practical application requirements and this study can provide investigators with a reference.

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