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Nondestructive measurement of total nitrogen in lettuce by integrating spectroscopy and computer vision

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ABSTRACT

This study was conducted to develop and assess a method of estimating the nitrogen (N) content of lettuce (*Lactuca sativa*) canopy using a combination of spectroscopy and computer vision for nondestructive N detection. In the experiment, 90 lettuce samples with five N treatments were collected for data acquisition by two different techniques. On the spectroscopy side, canopy spectral reflectance was measured in the wavelength range of 350 to 2500 nm at 1-nm increments. Four spectral intervals (376 variables) were selected by synergy interval partial least squares and were further reduced to 73 wavelength variables, chosen using a genetic algorithm applied to first-order derivatives of the canopy reflectance. On the computer vision side, 11 plant features were extracted from images, including top projected canopy area as a morphological feature; red, green, blue, hue, saturation, and intensity values as color features; and contrast, entropy, energy, and homogeneity as textural features. Next, principal component analysis was implemented on the spectral variables and on the image features, and extreme learning machine modeling was used to fuse the two kinds of data and construct a model. For the optimum model achieved, the root-mean-square error of prediction = 0.3231% and the correlation coefficient of prediction = 0.8864. This work demonstrates that integrating spectroscopy and computer vision with suitable efficient algorithms has high potential for use in the nondestructive measurement of N content in lettuce, considerably improving accuracy over that using a single sensor modality.

1. Introduction

Nitrogen (N) is a vital element in all organisms and is essential in various physiological processes of plants, where it is required continuously and in large amounts; it is a constituent of DNA, RNA, protein, chlorophyll, adenosine triphosphate, auxin (ATP), and cytokinins (Andrews et al., 2013; Zhang et al., 2013). A key concern for lettuce growers is to be able to verify plant N status early while N deficiency can still be remedied (Abdel-Rahman et al., 2010); thus, the early diagnosis of N deficiency plays a key role in the regulation of lettuce nutrition. However, the traditional process of N measurement has involved collecting samples at representative sites, pre-treating them, transporting them to the laboratory, analyzing them, and communicating the results back to the grower, which is a destructive, time-consuming, and inefficient procedure.

More recently, spectroscopy and computer vision, two commonly used nondestructive inspection techniques, have been widely employed to evaluate nutritional status expeditiously. Spectroscopy is an increasingly favored technique because of its rapidity, simplicity, and capacity to measure chemical characteristics, and it has been used to quantify the N content of plants by a number of researchers (Bajwa et al., 2010; Gislum et al., 2004; Miphokasap et al., 2012; Mitchell et al., 2012; Pacheco-Labrador et al., 2014; Padilla et al., 2014; Ramoelo et al., 2013; Ulissi et al., 2011). However, spectral data have high redundancy and collinearity and sometimes noise, which can reduce the estimation capability and computing efficiency of the model. Therefore, spectral transformation techniques are needed to enhance absorption features of biochemical constituents (Ramoelo et al., 2013), and wavelength selection methods are required to produce better prediction results with simpler models (Balabin and Smirnov, 2011). First-order derivative (FD) transformation can resolve problems such as multiple scattering of radiation due to sample geometry or surface roughness. By locating the positions of absorption features and inflection points on the spectra, it has proved to be a useful technique in estimating biochemical parameters such as

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chlorophyll and N content (Abdel-Rahman et al., 2010; Wang et al., 2013; Zhao et al., 2005). A continuum is a mathematical function used to isolate a particular absorption feature for analysis. With the continuum-removed (CR) approach, the reflectance spectra are normalized to enable comparison of individual absorption features of samples from a common baseline (Clark and Roush, 1984; Green and Craig, 1985; Kruse et al., 1985). Synergy interval partial least squares (siPLS) can help in searching all possible subinterval combinations to find the best model (Nørgaard et al., 2000). A genetic algorithm (GA) is an adaptive heuristic search algorithm that can be applied to spectral variable selection in combination with partial least squares (PLS) (Leardi, 2000). The siPLS algorithm can also be combined with GA, in an approach called GA-siPLS.

Computer vision has been applied in the visual evaluation of nutritional status by morphology, color, and texture (da Silva et al., 2014; Xu et al., 2011). Giacomelli et al. (1996, 1998) were able to determine nutrient stress in lettuce seedlings using a machine vision system that extracted the top projected leaf area. Ahmad and Reid (1996) compared red–green–blue (RGB), hue–saturation–intensity (HSI), and chromaticity coordinate color representations and their standards to evaluate the sensitivity of a machine vision system to detect color variations in stressed maize. Story et al. (2009, 2010) extracted the top projected canopy area, energy, entropy, and homogeneity and found them to be promising markers for the timely detection of calcium deficiency in lettuce; the methodology they developed was capable of identifying calcium deficiency one day prior to human visual detection.

However, it is difficult to fully assess N status using a single inspection technique such as spectroscopy or computer vision because one technique can gather only limited information. Clearly, spatial information cannot be extracted by spectroscopy, and chemical properties cannot be obtained using computer vision. In this study, we explore a new strategy for measurement of N content, one that integrates spectroscopy and computer vision. The intent is to take advantage of the strengths of the two techniques, spectroscopy performing a local measurement of inner chemical properties, computer vision performing a global assessment of external physical properties, and combine these two measurements in a way that improves N content assessment. The specific objectives of this study are (1) to identify the most suitable wavelengths for quantifying N; (2) to extract the morphology, color, and texture features from canopy images; and (3) to merge data from the two sensor modalities to assess the N content of lettuce and compare the performance of the combined model to that of the two individual models.

2. Materials and methods

2.1. Sample preparation

The experimental materials were lettuce (*Lactuca sativa*, from Woshu Seeds Co. Ltd., Nanjing, China). The experiment was performed in the greenhouse at Jiangsu University in China (32.11N, 119.27E). All the plants investigated were transplanted during the period of five true leaves and grown under non-soil conditions from May to June 2012. The Yamasaki lettuce recipe was used for lettuce growth. The composition was: $Ca(NO_3)_2{\cdot}4H_2O$, 236 mg/L; KNO_3, 404 mg/L; $NH_4H_2PO_4$, 57 mg/L; $MgSO_4{\cdot}7H_2O$, 123 mg/L; Fe-EDTA, 16 mg/L; $MnCl_2{\cdot}4H_2O$, 1.2 mg/L; H_3BO_3, 0.72 mg/L; $ZnSO_4{\cdot}4H_2O$, 0.09 mg/L; $CuSO_4{\cdot}5H_2O$, 0.04 mg/L; and $(NO_4)_2Mo_7O_4$, 0.01 mg/L. In this solution, NO_3^- concentration was 6 mmol/L, and NH_4^+ concentration was 0.5 mmol/L. For our study, we used five treatments having different levels of N, containing, respectively: (1) NO_3^- 1.5 mmol/L and NH_4^+ 0.125 mmol/L, (2) NO_3^- 3 mmol/L and NH_4^+ 0.25 mmol/L, (3) NO_3^- 4.5 mmol/L and NH_4^+ 0.375 mmol/L, (4) NO_3^- 6 mmol/L and NH_4^+ 0.5 mmol/L, and (5) NO_3^- 7.5 mmol/L and NH_4^+ 0.625 mmol/L. Each group of lettuce roots was kept on its fixed nutrient solution content by a self-developed timed irrigation and collection system.

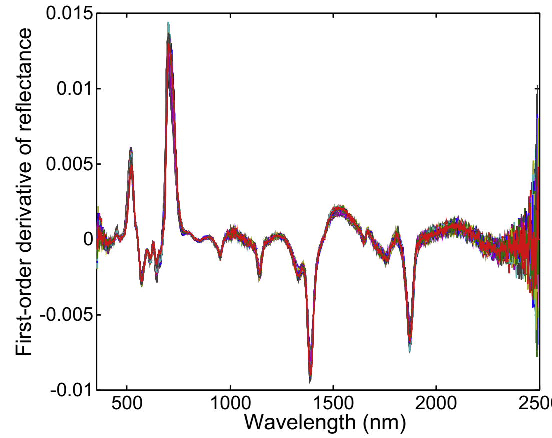

Fig. 1. First-order derivative of reflectance.

2.2. Spectral data acquisition and preprocessing

Canopy reflectance was acquired with a FieldSpec 3 spectroradiometer [Analytical Spectral Devices (ASD), Boulder, CO, USA] that provides measurements in the spectral range of 350–2500 nm. The built-in spectral resolution of data output from the ASD operating system is 1 nm along the whole spectrum. The spectra of all samples were measured in an opaque closed box with a 50-W halogen lamp as the light source. The diameter of the light spot was about 9 cm, from a fiber optic cable with a 25° field of view. Prior to the first scan, a calibration was performed by taking dark current and using a standard white Spectralon panel (Labsphere Inc., USA) with nearly 100% reflectance. Five spectral measurements were taken for each sample in order to attain a reduction in noise by averaging the spectra.

To eliminate multiple scattering, high-frequency noise, and other interferences that can reduce the performance of the model, it is necessary to apply an appropriate mathematical technique. In our analysis, the first step was to smooth the data using a nine-point weighted moving average to remove noise. Then, FD spectra (Fig. 1) and CR spectra (Fig. 2) were calculated from the smoothed reflectance. Both FD and CR preprocessing methods were

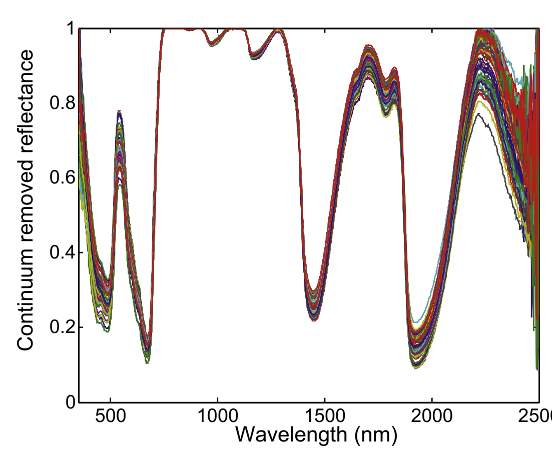

Fig. 2. Continuum-removed reflectance.