Rapid identification of mulberry leaf pests based on near infrared hyperspectral imaging

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A paper from the Proceedings of the
14th International Conference on Precision Agriculture
June 24 – June 27, 2018
Montreal, Quebec, Canada

Abstract. *As one of the most common mulberry pests, Diaphania pyloalis Walker (Lepidoptera: Pyralididae) has occurred and damaged in the main sericulture areas of China. Naked eye observation, the most dominating method identifying the damage of Diaphania pyloalis, is time-wasting and labor consuming. In order to improve the identification and diagnosis efficiency and avoid the massive outbreak of Diaphania pyloalis, near infrared (NIR) hyperspectral imaging technology combined with partial least discriminant analysis (PLS-DA) and Successive projections algorithm (SPA) algorithm was applied to establish a fast and nondestructive detection method of Diaphania pyloalis larva. Hyperspectral images of samples were collected and corresponding spectra data was extracted, then classification models were established. Results showed that the mean value of the correct rate of calibration and prediction (M_CR) of PLS-DA model with full variables was 76.65%, nevertheless, the absolute difference between the correct rate of calibration and prediction (AB_CR) value was 31.55%. After variable selection calculation, the AB_CR value was reduced to 2.77% based on SPA-PLS-DA model with 9 selected variables, it showed that the robustness of model was improved. In conclusion, hyperspectral imaging coupled with chemometrics method showed a certain potential in the rapid detection of Diaphania*

pyloalis larvae on mulberry leaves.

Keywords.

Near infrared hyperspectral imaging, PLS-DA, SPA, mulberry leaf, Diaphania pyloalis larva.

Introduction

Mulberry (*Morus alba L.*), a short-lived, fast-growing perennial plant, is belonging to the genus Morus and the family Moraceae. Mulberry is widely grow in different climate zones, not only the temperate to subtropical regions in the northern hemisphere but also the tropical regions in the southern hemisphere. There are approximately 24 members in the genus Morus and one subspecies which have more than 100 varieties as far as we all know (Ercisli and Orhan 2007), and the most commonly planting species of which are white mulberry (*Morus alba L.*), black mulberry (*Morus nigra L.*) and red mulberry (*Morus rubra L.*) (Ercisli and Orhan 2008). Originated from China and meanwhile widely naturalized and planted elsewhere, white mulberry is dominant among the three major mulberry species.

Mulberry planting and silkworm rearing is a traditional industry for silk production in China and has history of thousands of years. Nowadays, mulberry is planting as an economical woody plant in most parts of China. The economic value of mulberry is mainly reflected in two aspects: mulberry fruits and mulberry leaves which are widely used as traditional Chinese medicine, fresh fruit, food, feed and horticultural crops (Khalifa et al. 2018). Mulberry fruit is a very delicious and popular fruit in China, and it is rich in nutrients. Studies have shown that mulberry fruits are a remarkable source of polyphenols (Khalifa et al. 2018). In addition to being a food item, in the main mulberry growing regions, especially in China and India, mulberry trees are used for its leaves as a feed source for silkworm (*Bombyx mori L.*) rearing. On the other hand, as the sole feed for silkworm, the quality of mulberry leaves directly determines the economic benefits of sericulture. Therefore, the management of mulberry cultivation focuses more on the improvement of the foliage quality and yield in these countries (Vijayan et al. 1997). In the past ten years, despite the expansion of the mulberry planting area, the output of mulberry leaves has declined slightly due to the inappropriate management in China. As part of the mulberry production management, pest control has gradually received interest and attention of the mulberry famers.

Diaphania pyloalis Walker (Lepidoptera: Pyralididae) as one of the most common mulberry pests, is widely found in mulberry-planting areas and causes serious loss of mori folium yield (Zhu et al. 2013). Mulberry leaf is also the only food of *Diaphania pyloalis*, hence, the existence of *Diaphania pyloalis* forms a great competition for silkworm. Before 1990s, *Diaphania pyloalis* was not the main pest of mulberry trees. The extensive use of pesticides in mulberry management killed pests and also killed the natural enemies of *Diaphania pyloalis*, it gradually became one of the major pests in mulberry fields. It was reported that the outbroke of *Diaphania pyloalis* from 1997 to 1999 had damaged mulberry in an area of 300 km^2 in the areas of northern Zhejiang province, China (Bai et al. 2002). Nowadays, *Diaphania pyloalis* poses a threat in the main mulberry-growing areas of China and also other regions such as Japan, Indian and Southeast Asia. Therefore, it is necessary for developing a rapid detection method for *Diaphania pyloalis* pests to ensure the quality of mulberry production for silkworm rearing.

Naked eyes observation is a commonly used method of pest detection. However, subjective observation is labor-consuming, inefficient and costly. Moreover, the small *Diaphania pyloalis* larvae often have similar colors to the mulberry leaves, it is very usual for missing larvae and error-prone due to visual fatigue based on naked eyes observation. In view of the above situation, computer vision technique was developed to provide objective inspection for pests in low-cost ways (Cheng et al. 2017; Liu et al. 2016). The biggest difference between computer vision and visual inspection is that the machine will never be tired and will not be influenced by human subjective factors. Nevertheless, traditional computer vision works like the human eyes which are sensitive to the three primary colors-red, green and blue (RGB), and uses external RGB color cameras with three filters centered at RGB wavelengths to obtain image information (Lorente et al. 2012). Therefore, the images acquired by the computer vision based on the RGB color camera is very close in color to the actual image obtained by the human eyes (Zhang et al. 2014). Because the larvae and the leaves are similar in color, it is difficult to distinguish pests and leaves by naked eyes observation or computer vision.

As a development of computer vision technique, hyperspectral imaging (HSI) is a potential tool for non-contact and rapid acquisition of surface and inherent information of the specimen in agriculture and food products. In recent years, the application of HSI has ranged from the quality inspection of food (Elmasry et al. 2011; Huang et al. 2017) and agricultural products (Li et al. 2011; C. Zhang et al. 2015) to the detection of plant diseases (Kong et al. 2018; Jaillais et al. 2015; Fan et al. 2017) and insect pests (Saranwong et al. 2010; Lu and Ariana 2013; Ma et al. 2014). HSI is the integration of traditional spectroscopy with traditional imaging techniques. Traditional spectroscopy is mainly used to measure the optical properties of materials but lack the spatial distribution of the features. On the other hand, spatial distribution of traits of the specimen can be requested by conventional imaging system or machine vision. By combining the core advantages of spectroscopy and imaging, hyperspectral imaging technology enables simultaneous acquisition of spectral and spatial distribution information of the sample. The two types of information obtained by HSI are integrated to form a three-dimensional (3D) data cube composing of vector pixels that is including one spectral dimension (***λ*** wavelengths) and two-dimensional spatial information (***x*** rows and ***y*** columns) (Wu and Sun 2013). Considering the characteristics of HSI and the previous studies, it is worthwhile to identify pests and mulberry leaves and to locate the spatial position of pests using the abundant information in the 3D hypercube obtained by HSI. Nevertheless, there are few reported study on the rapid detection of *Diaphania pyloalis* pests on mulberry leaves using HSI techniques.

In this work, near-infrared (NIR) hyperspectral reflectance imaging technology coupling with chemometrics methods was used to identify pests from mulberry leaves. The success of this work is conducive to strengthen the management of mulberry trees, provide high-quality and high-yield mulberry leaves for the silkworm, and promote the healthy and long-term development of the sericulture industry. The specific content of this study included: (1) acquiring the hyperspectral images of the samples in the spectral range of NIR (900- 1700 nm), (2) extracting the characteristic spectra of *Diaphania pyloalis* larvae and mulberry leaves, (3) establishing classification model of *Diaphania pyloalis* larvae and mulberry leaves, and (4) identifying a few useful wavelengths for the classification.

Materials and methods

Sample preparation

Mulberry leaves without any visual defects and *Diaphania pyloalis* larvae were collected at an experimental mulberry planting base in Zijingang campus, Zhejiang University (Hangzhou, Zhejiang province, China) on Oct. 24, 2017. The collected samples were stored without any processing and hyperspectral image acquisition was performed within two hours to ensure that the hyperspectral image information was close to that of the fresh leaves in mulberry trees.

Hyperspectral imaging system and data acquisition

Hyperspectral imaging instruments are the foundation and important medium for obtaining reliable hyperspectral images with high quality. The hyperspectral imaging system used in this work was mainly composed of two parts, namely spectral acquisition unit and control unit. The spectral acquisition unit was the most important component of the HSI system, and mainly made up of three components: a camera, an imaging spectrograph which measured hyperspectral images in 900-1700 nm and two adjustable quartz tungsten halogen lamps with power of 150 w. The spectral acquisition unit was installed in a black box which was closed during data collection to avoid interference from external light. The control unit consisted of a conveyer platform, a stepper motor and a computer. During images collection, samples were placed on the conveyer platform which were controlled by software on the computer. The approaches to acquire hyperspectral images of samples was called line-scanning, and the image sensing mode was reflectance.

Processing of hyperspectral images

After the data acquisition was complete, raw hyperspectral images were collected. The raw image collected by hyperspectral imaging was the detector signal intensity. Therefore, the raw images needed to be corrected with the help of the image of the white and dark reference collected in the same condition based on the below equation (Sun et al. 2017).

$$R = \frac{I_R - I_D}{I_W - I_D} \times 100\% \qquad (1)$$

where R is the corrected image, I_R is the raw hyperspectral image, I_D is the dark image with about 0% reflectance acquired with the camera lens covered by its opaque cap and lights turned off, and I_W is the white reference image collected based on Teflon white board which had stable, uniform and high reflectance standard with a reflectance close to 99% under the same experiment condition. The role of the dark reference was to eliminate the dark current influence of the camera sensor. The corrected hyperspectral image was the basis for follow-up work.

Data analysis

Spectral data extraction

A commercially available data processing software tools called Environment for Visualizing Images (ENVI) software (Research Systems Inc., Boulder Co., USA) was used to extract the spectral data of samples. There was a function called region of interests (ROI) in the ENVI v4.6 software. The objective of this function was located the target regions and divided the images for further spectral and textural feature extraction. The ROIs of mulberry leaves and *Diaphania pyloalis* larvae were identified and then the spectral data of mulberry leaves and *Diaphania pyloalis* larvae were extracted within the corresponding ROIs.

Due to the differences between the spectra of the samples, none of the ROI could represent a class of sample. Therefore, mean spectrum of each class ROI was calculated to represent the spectra of this ROIs. This work was finished with the help of MATLAB v2015b (The Math-Works Inc., Natick, MA, USA). The mean spectral data was then used to establish models.

Multivariate classification model

Due to the huge amount of spectral information obtained in the hyperspectral images, it was necessary to extract useful feature texture information through multivariate analysis and establish the relationship between sample hyperspectral data and sample attributes. Supervised qualitative classification algorithm partial least discriminant analysis (PLS-DA) was applied by MATLAB v2015b (The Math-Works Inc., Natick, MA, USA) in this work.

PLS-DA was a discriminant algorithm developed on the basis of partial least squares regression (PLSR) which was widely used in spectra analysis (Frank et al. 1983). The two algorithms PLS-DA and PLSR have the same basic calculation principle which was established model counting on the extracted latent variables.

PLSR was a regression modeling method which absorbed the advantages of principal component analysis and multiple linear regression analysis for multiple dependent variables (***Y***) and multiple independent variables (***X***). In the process of establishing regression relationship, the algorithm not only considered extracting the principal components of ***Y*** (sample attributes) and ***X*** (sample hyperspectral data) as much as possible, but also considered maximizing the correlation between principal components extracted from sample attributes and sample hyperspectral data, respectively. Different from PLSR algorithm, the dependent variables ***Y*** in PLS-DA algorithm was not represented the desired attributes of samples, it was a series of dummy integer numbers set to represent the classes of samples. In other words, the spectral data of the samples (***X***) were established regression relationships with the specific numbers (***Y***). Nevertheless, the predicted value based on the established regression relationship was a real number. Therefore, a cut-off value was chose to determine the class of the predicted value. Usually, the cut-off value was selected as 0.5 (Kong et al. 2013). The PLS-DA algorithm had proven to be an effective method of model establishment in many applications (Talens et al. 2013; Andre 2003).

In this work, three classes mean spectral data was extracted from the ROIs which were leaf vein, healthy leaf mesophyll and *Diaphania pyloalis* larva. Consecutive natural numbers 1, 2 and 3 were chose to represent the above three types of data respectively and then modeled with the corresponding mean spectral data using PLS-DA algorithm.

Variable selection

Hyperspectral imaging provides extremely rich spectral and spatial information of samples, which was an advantage of this technique. Nevertheless, the information of most wavelengths contained a lot of unrelated information while other wavelengths had low signal-to-noise ratio (SNR). In most cases, these wavelengths did not make much contribution to the establishment of the model. In addition, the existence of the collinearity between variables in the model affected the singularity of the matrix, and that had an influence on the results of modeling (Zou et al. 2010). Therefore, it was necessary to eliminate the irrelevant variables to simplify modeling calculations and improve the accuracy and stability of the model (Wu et al. 2008; Wu et al. 2009).

The variable selection was used to select useful wavelengths associated with the quality attribute of samples to reduce the errors resulting from the qualitative discrimination and improve the robustness of model. Successive projections algorithm (SPA) was a relatively complex and promising variable selection method designed to solve the collinearity problems by selecting variables with minimal redundancy (Araújo et al. 2001). In this work, SPA was applied to select the optimal wavelengths that did not suffer from redundancy and contributed most to the classification of samples. The SPA calculation process usually consists of two steps. Step one, starting from the first wavelength, a series of simple projection operation was performed on the column of the spectral matrix to build candidate subsets of variables with minimal collinearity. Step two, the candidate subsets were evaluated according to the predicted performance of the obtained multiple linear regression model (Wu et al. 2013).

Model evaluation

In the qualitative analysis of hyperspectral data, multivariate data was divided into a calibration set and a prediction set. The data of calibration set was trained to establish a calibration model, and then the data of prediction set was used to evaluate the performance of the model. In this study, the performance of the classification model was evaluated in terms of correct rate of calibration (CRC), correct rate of prediction (CRP), mean value of the correct rate of calibration and prediction (M_CR), and the absolute difference between the correct rate of calibration and prediction (AB_CR). The CR represented the number of correct prediction of samples as a percentage of the total number of samples, and AB_CR represented the robustness of the model. In general, a good model should have higher CR value and lower AB_CR value.

Results and Discussion

Results

Diaphania pyloalis larva nibbled mulberry leaves, causing a decline in the quality and quantity of mulberry leaves and posed a threat on the management of mulberry production. The number of larva on the leaves was an important indicator for evaluating the degree of the damages caused by the pests. Moreover, the detection of larvae was an important basis for taking subsequent prevention and control measures. Therefore, it was significant to identify *Diaphania pyloalis* larvae from the mulberry leaves. The classification results for leaf vein, healthy leaf mesophyll and *Diaphania pyloalis* larva based on full variables and selected variables were shown in Table 1.

Table 1. Results of classification models for identifying leaf vein, healthy leaf mesophyll and Diaphania pyloalis larva based on the full variables and selected variables.

Model	Variable selection	Variable number	Calibration	CRC[a]	CRP[b]	M_CR[c]	AB_CR[d]
I[e]	/	256	PLS-DA	92.42%	60.87%	76.65%	31.55%
II[f]	SPA	9	PLS-DA	63.64%	60.87%	62.25%	2.77%

[a] correct rate of calibration, [b] correct rate of prediction, [c] mean value of the correct rate of calibration and prediction, [d] the absolute difference between the correct rate of calibration and prediction, [e] Model I based on full variables, [f] Model II based on selected variables

When Model I was considered, the full wavelengths spectra data contained a total 256 variables based on NIR hyperspectral imaging. Relationship between spectral data and classification values established using the PLS-DA algorithm, the overall CRC value was 92.42%, nevertheless, the overall CRP value was 60.87%, and the M_CR value was 76.65%. The AB_CR value that represented the robustness of the model was as high as 31.55%.

Model II was established based on variable selection SPA algorithm. The overall CRC value was 63.64%, the CRP value was 60.87%, and the M_CR value was 62.25%. The AB_CR value was only 2.77% and the selected variables was reduced to 9.

Discussion

Mulberry leaves with high quality and quantity are required, no matter it is used as a feed for rearing silkworms or as a raw material for food processing. Mulberry leaf pests are a major factor causing the decline of mulberry quality and yield. Nowadays, *Diaphania pyloalis* is the main pests that threatens the production of mulberry leaves in China. In this work, near infrared hyperspectral imaging technique in the range of 900-1700 nm was applied to detect and identify *Diaphania pyloalis* larvae. In the classification Model I, the CRC value was high while the CRP was low (92.42% vs. 60.87%), the AB_CR was 31.55%. The results showed that Model I based on full variables had poor robustness. The most likely reason for this situation was the high degree of collinearity among the variables in the spectral data, resulting in overfitting of the calibration model. Therefore, the SPA algorithm was used to remove the redundant variables to solve the collinearity problem between variables and improve the robustness of model. The results after the variable selection calculation was shown in classification Model II. As we can see, the number of input variables was only 9 and it was 96.48% lower than the number of full variables which was 256. The CRC value and CRP value of the model based on the selected 9 variables was 63.64% and 60.84% respectively. The AB_CR value of SPA-PLS-DA model was only 2.77%, which was a 28.78% decrease compared to the AB_CR value of the PLS-DA model with full variables. It showed that the SPA-PLS-DA model had a good robustness and variable selection SPA algorithm could reduce the variables and solve the collinearity problem between variables in the PLS-DA model with full variables.

Conclusion

Near infrared hyperspectral imaging technique was carried out to evaluate the feasibility of rapidly identifying *Diaphania pyloalis* larvae from mulberry leaves in this study. Classification model between mulberry leaves and *Diaphania pyloalis* larvae was established. The M_CR value was 76.65% based on PLS-DA model with full variables. Nevertheless, the AB_CR value that represented the robustness of model was 31.55%. Then, classification model based on variable selection was established, a total of 9 variables was selected. The M_CR value was 62.25%, and the AB_CR value was reduced to 2.77% which showed that the model had a good robustness. As a preliminary study, NIR hyperspectral imaging was first used to identify *Diaphania pyloalis* larva on mulberry leaves, and the results showed a potential of this technique and would help ensure the quality and yield of mulberry leaves.

Acknowledgements

This work was supported by the National Key R&D Program of China (2017YFD0401302 and 2016YFD0700304), the Fundamental Research Funds for the Central Universities (2017QNA6025); National Natural Science Foundation of China (31701654); Science and Technology Plan Project in Zhejiang Province (2017C32013); and Huzhou Public Welfare Technology Application Research Project (2015GZ12).

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