Non-destructive prediction of total soluble solids, titratable acidity and maturity index of limes by near infrared hyperspectral imaging

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ABSTRACT

This study was implemented for non-destructive prediction of total soluble solids (TSS), titratable acidity (TA) and calculation of TSS/TA as a measure of maturity index in intact limes using laboratory-based push-broom hyperspectral imaging (HSI) in reflectance mode in the range of 929–1671 nm. Limes were scanned by the HSI system in order to develop calibration models for predicting TSS, TA and TSS/TA using partial least square regression (PLSR). Original spectra obtained optimal conditions for establishing the models for TSS and TA while smoothing spectra for TSS/TA. The accuracy of the models for TSS, TA and TSS/TA provided coefficient of determination of prediction (R_p^2) of 0.838, 0.694 and 0.775, respectively and root mean square errors of prediction (RMSEP) of 0.237%, 0.288% and 0.049, respectively. Image processing algorithms were then built up by interpreting predictive values, from the models, to colors in each pixel of the images. The predictive visualization of TSS, TA and TSS/TA in all portions of the limes based on a color scale was presented. The results showed that the HSI technique has the capability of predicting TSS, TA and TSS/TA of intact limes non-destructively and the results could be visualized by different colors of the predictive images.

1. Introduction

Limes (*Citrus aurantifolia* Swingle) are an important commercial citrus fruit and their composition varies with cultivar, climate, rootstock and cultural practices. They are marketed and utilized mainly as fresh fruit but a considerable proportion is marketed as processed products such as lime juice. The total world production of limes and lemons in 2016/17 was 7.3 million metric tons of which 2 million metric tons were used for processing (United States Department of Agriculture, 2017). A non-destructive method for evaluating the quality of limes would be valuable in grading the fruit to provide consistency in the processed product as well as grading for the fresh market. It would also have application in other citrus fruit. The measurement of TSS and TA, besides being a measure of the quality of the fruit, enables the calculation of the TSS to TA ratio, which is a common measure of maturity in non-climacteric fruit (Reid, 2002). Fruit maturity is an important criterion of fruit quality both in the packing house for the fresh market and in the factory for processing.

It has been reported that near infrared (NIR) spectroscopy and image feature extraction techniques could be used to evaluate, non-destructively, qualities of various intact fruit and vegetable (Walsh et al., 2004; Zheng et al., 2006). HSI represents the spectral data at each pixel of an image, which has not been achieved with either conventional imaging or spectral spectroscopy individually. It is a powerful technique which has been developed as a logical extension of both near infrared spectroscopy (NIRs) and computer vision technology for rapid, non-destructive determination of various features of agricultural products (Chen et al., 2002).

For application, it is necessary to perform a calibration by the determination of the relationship between the actual chemical composition of the fruit samples and their spectra to build the models to predict, non-destructively, the desired dependent variables of the samples. Specifically, chemometrics is used, which is the combination of the application of mathematical and statistical methods to break down complex spectral data into applicable structures that can improve the understanding of the data from the samples (Alamar et al., 2016). PLSR is frequently applied by using the information of both the independent and the dependent variables. It has been effectively used in analyzing spectral data for dimensionality reduction and extracting the useful information from NIR data in order to develop the calibration model (Fernandez Pierna et al., 2003). It has resulted in considerable interest on the application of NIR spectroscopy for monitoring and evaluating quality of fruit specifically and for the fruit processing industry. There are many available commercial conventional NIR spectrometers which are used for measurement. Only spectral information from a specific point of the scan on the sample is acquired and used as a representative.

The average value of the composition over the entire sample, which is related to organic molecules that contain C—H, C—O, O—H and C—C bonds, is provided continuously for analysis. There are several studies that demonstrate the possibility of using conventional NIR spectroscopy to identify external and internal qualities of citrus fruit (Umezuruike et al., 2011). These include physical quality parameters of oranges and mandarins (Torres et al., 2017); fungal infections in mandarins (Lorente et al., 2015); sucrose, glucose and fructose in oranges (Tewari et al., 2008); citrus canker in grapefruits (Balasundaram et al., 2009) and TSS and TA in Valencia oranges (Ncama et al., 2017). Lorente et al. (2012) showed that the spectra of each pixel, which provide the spatial and spectral information, can be acquired by HSI. The multivariate model can be established to predict every single pixel of HSI and transfer them to physiochemical images (Burger and Geladi, 2006). The acquired images show the spatial distribution of the chemical composition of the tested sample. For fruit samples, the physicochemical images are usually created to visualize their internal quality by presenting quantitative spatial distribution of the compositions and their relative concentrations. Other studies on the quality assessment of citrus fruit using HSI include the detection of early symptoms of decay in navel oranges (Li et al., 2016), detection of green mould in mandarins (Folch-Fortuny et al., 2016) and detection of thrips on the green peel of citrus (Chunwang et al., 2014). The objective of this research was to test whether it is possible to relate HSI measurements to TSS, TA and the calculation of TSS/TA of intact limes and to visualize these measurements by color distribution images. This could provide a simple system that would be applied in packing houses for fresh limes and factories for processed limes.

2. Materials and methods

2.1. Lime samples

A total of 100 limes (cv. Paan), which were selected for color (fully green) and size uniformity (4.0 ± 0.3 mm), were collected at the wholesale market in the KhonKaen province of Thailand. Samples were stored at 25 °C in an air conditioned room for about 24 h before measurement. After scanning by HSI, fresh juice was extracted by individually squeezing each lime. TSS was measured using a digital refractometer (PR101, Palette Series, Atago Co., Ltd., Tokyo, Japan) and TA was determined by titration of a 10 mL sample and 50 mL distilled water with 0.1 M sodium hydroxide, using phenolphthalein indicator, until the pink end-point. TA was calculated by the Eq. (1) as outlined in Ncama et al. (2017) and was expressed as the citric acid content (%) of the sample.

$$TA\,(\%\ citric\ acid) = \frac{(0.0064)(volume\ of\ NaOH\ in\ mL)(100)}{10} \quad (1)$$

2.2. NIR-HSI system

Each lime was image scanned individually in the line-scan push broom Sisu CHEMA (Fig. 1), which consisted of an imaging spectrograph (Imspector N17E, Specim, Spectral Imaging Ltd, Oulu, Finland), an electron-multiplying charge-coupled device (EMCCD) camera, an adjustable slit, two reflectors, an illumination unit including two halogen lamps (Lowel Light Inc., NY, USA), an OLES 22.5 fore objectives lenses, fiber optic, a sample stage, an internal white reference, a linear positioning stage, a power supply unit combined with a PC acquisition supported with SpectralCube data acquisition software from Spectral Imaging Ltd. (Oulu, Finland). The spectrograph consisted of a prism-grating-prism arrangement that dispersed the reflected light for an optical spectral resolution of 5 nm. The camera was attached to the image spectrograph SPECIM Inspector N17E of the spectral range 897–1752 nm with 256 spectral bands and 320 × 256 (spatial x spectral) pixels. The lighting system came from two line lights (halogen
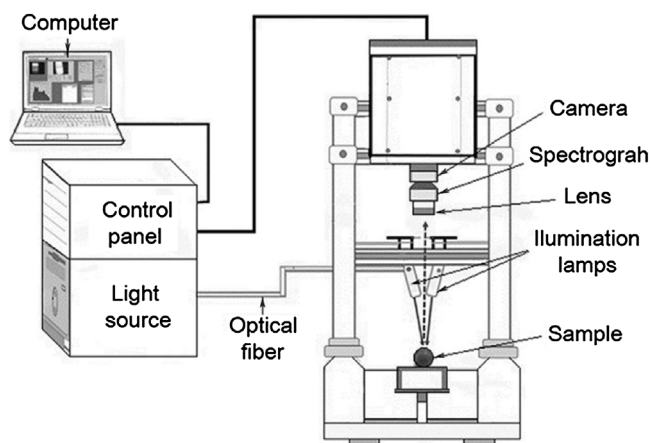

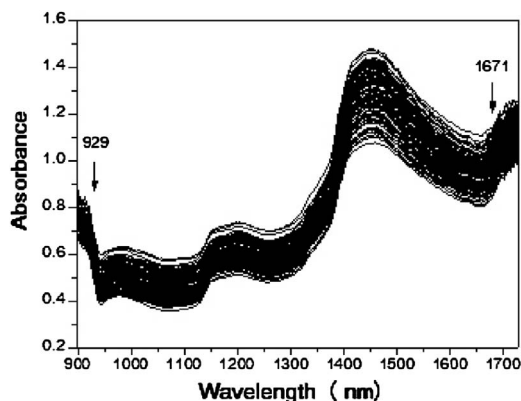**Fig. 1.** Schematic representation of the HSI system.

Fig. 2. The original absorbance spectra of limes in the range of 896–1731 nm.
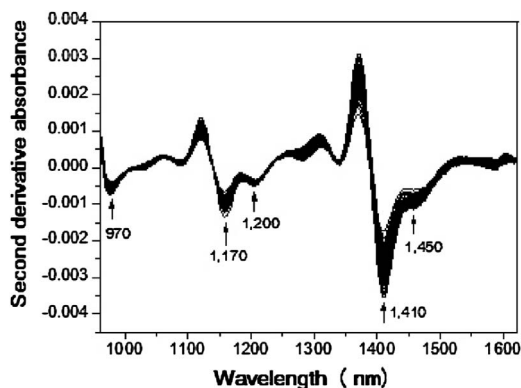

Fig. 3. The second derivative absorbance spectra of limes in the range of 929–1671 nm.

lights, 1400 nm notch filter) that were arranged to illuminate the IFOV (100–31 mm lens). The light that passed the objective lens and the entrance slit of spectrograph was dispersed into its NIR spectrum. The reflected light from the IFOV passed through the slit and was dispersed by a prism device and projected into the EMCCD sensor. The particular speed was precisely calculated to fit the predetermined camera exposure time or integration time (1.5 ms) and scanning frequency (45 frames per second) to provide the same vertical and horizontal spatial resolution. The whole system was put inside a dark cubicle to avoid any stray lighting in the room that could have affected the recorded reflectance from the sample. In addition, the whole image acquisition process was controlled and recorded by a computer (64-bit Java) supported by using the ChemaDAQ data acquisition software. (Specim, Spectral Imaging Ltd, Oulu, Finland).

Three main procedures were applied on HSI in this study: (1) image