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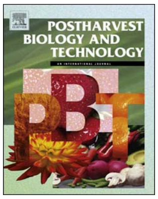

# Discriminating hidden bruises in loquat by attenuation coefficients estimated from optical coherence tomography images

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## ABSTRACT

Hidden bruising is considered as one of the difficulties during loquat quality control, which has made an impact on postharvest quality and storability. Optical coherence tomography (OCT) is used as a non-destructive method that acquires 2D images of the sub-surface structure of the loquat. After OCT image processing, the attenuation coefficients ($\mu_t$) of the regions of interests are fitted to quantitatively analyze the characteristics of loquat tissue through using a confocal signal scattering model from OCT A-scan signals. In this experiment, a total of 60 loquat samples are investigated, with the non-bruised tissue having a large $\mu_t$ with a mean value of 1910 $\mathrm{m}^{-1}$ (std: ± 160). However, bruised groups without any storage time and after storage for 12 h showed lower $\mu_t$ with a mean value of 1190 $\mathrm{m}^{-1}$ (std: ± 230) and 1020 $\mathrm{m}^{-1}$ (std: ± 190) respectively. The $\mu_t$ value is sensitive to chemical and structural changes in loquat tissue and can also be used in hidden bruise discrimination of loquat.

## 1. Introduction

Loquat is a popular fruit produced in subtropical regions. The fruit has a sweet taste and is very nutritious (Moreira et al., 2008). During or after the harvest, mechanical damage becomes the major reason for the premature decay of fresh fruit and a short shelf life. During the process of picking, packing and transportation, the fruit is subjected to stress, resulting in permanent damage to the sarcocarp (Wang et al., 2016). This damage leads to partial rotting of the flesh, which can contaminates the rest of the fruit (Lin et al., 2016). This situation not only leads to imperfect appearance and dissatisfied consumers, but also promotes disease cross-contamination between fruits in one batch. Therefore, it is necessary to establish a non-destructive discrimination method for hidden bruises in fresh loquat fruit.

In the present study, we mainly focus on the early bruises of the loquat. As the bruised flesh decomposes, the surface of the fruit appears brown. Based on the color characteristics, it is effortless to discriminate bruised ones using color-based methods (Zhang et al., 2015). A certain amount of fruit grading lines are based on computer vision technique (Zhang et al., 2014), so the challenge is to discriminate hidden bruises in the early stage of damage before any color change occurs. Recently, spectroscopic techniques have been successfully applied in the fast and non-destructive determination of loquat internal quality. Visible near infrared spectroscopy and near infrared hyperspectral imaging are two typical examples of these techniques which have been applied to detect soluble solids content in loquat (Yu et al., 2014; Fu et al., 2007). However, the visible-near infrared spectroscopy relies on the absorption of flesh, and its accuracy is vulnerable to the variety, size, origin and habitat of the loquat. Besides, hyperspectral imaging provides the spatial distribution of absorption or reflection information, which is more useful than the information provided by visible-near infrared spectroscopy. In any case, spectroscopy techniques need to use chemometrics method for pretreatment, wavelength selection, building calibration models, and feature selection (Lourenço et al., 2012). Although these methods have experienced rapid development, quality parameters estimated from them reflect the near surface situation. Particularly in early bruise discrimination, characteristics of the flesh under peel change and need to be closely monitored. It is meaningful to establish a hidden bruises discrimination method considering both spatial and depth information, which can thus make the improvement of prediction accuracy and stability.

Optical coherence tomography (OCT) can obtain the sub-face sectional information from low light coherence interferometry, which has advantages of being non-destructive, contactless and being high resolution in nature (Adhi and Duker, 2013). Utilizing the galvanometer scanner, the OCT device acquires three dimensional images with the consideration of depth resolved data under the peel surface with micrometer resolution. The OCT device can be categorized into two groups respectively, time domain OCT (TD-OCT) and spectral-domain OCT (SD-OCT) (Hahn et al., 2015). SD-OCT uses subsequent

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fast Fourier transformation to execute the depth scans, instead of shifting reference mirror in TD-OCT, and reaches the high acquisition speed as required in online measurement. Increasingly, SD-OCT applications are used in clinical practice, such as procedures as the assessment of ambiguous angiographic lesions and identification of vulnerable plaques (Prati et al., 2012).

The SD-OCT has been gradually introduced into agricultural research. Microstructure of apple peel and near skin cellular have been characterized by OCT images (Verboven et al., 2013). Visualization of plants and plant cells for the quantitative study of the hull thickness of lupin seeds (Lee et al., 2012), disease detection in melon seeds and apple leaves (Wijesinghe et al., 2016), detection of defects, rotting, and diseases in onions are also proposed OCT applications in agriculture (Meglinski et al., 2010). Admittedly, studies have not been done regarding discrimination of hidden bruise in loquat flesh, however, we believe that SD-OCT could be an effective tool for monitoring the sub-surface changes of loquat.

The loquat injury or damage causes changes in the micro-structure and composition of tissue that influences the size and shape of cells (Aghdam and Bodbodak, 2014). After receiving scattered and reflected photons, the SD-OCT image corresponds to these changes. The optical properties are considered as a powerful tool to distinguish between healthy and bruised tissues. Loquat flesh causes light to be both scattered and absorbed when light penetrates tissue (Lorente et al., 2015). The total attenuation coefficients ($\mu_t$), the sum of the two interactions of scattering and absorption, can be fitted from A-scan measurements (Vermeer et al., 2014). Because the OCT image obtains deeper information of tissue, it is more applicable to monitor $\mu_t$ changes of flesh, and it also has considerable potential to overcome the limitation wherein flesh construction and constituent changes have not caused any peel color variance in the early bruising stage. Flesh bruising leads to changes in the soluble solids content, titratable acidity and firmness, as well as other parameters which may affect the results of $\mu_t$ estimation.

Loquat OCT image provides contrast differences based on reflectivity of loquat flesh, but the application of contrast for discrimination has a limitation when there are small difference in contrast. However, the estimation of $\mu_t$ supplies additional quantitative indicators. At the same time, single scattering and multiple scattering models are available to describe the relationship between $\mu_t$ and OCT signals in the region of interest (ROI) (Van der Meer et al., 2005; Faber et al., 2004).

This work aims at investigating whether OCT can be used for non-destructively identifying hidden bruising in loquat flesh. The OCT technique visualizes and characterizes the sub-surface structures of loquats, and allows the subsequent estimation of $\mu_t$ in the ROI on the basis of fitting theoretical models.

## 2. Materials and methods

### 2.1. Loquat samples and treatment

A total of 60 loquat fruit were harvested in May 2015 in Thang xi town, which is one of the main producing areas in China. All the loquat fruit were of the variety of 'Baisha', which was popular among customers.

A mechanical treatment was applied to simulate hidden bruising in the loquat samples. The self-made device for applying mechanical force consisted of a 0.02 m diameter stainless steel cylinder and 1.8 cm diameter steel ball weighting $1.68 \times 10^{-4}$ kg. The hidden bruise in the loquat flesh was formed by dropping the steel ball through the cylinder onto the loquat from a height of 0.2 m. This method was similar to that reported by Testoni and Grassi (1995). After impact, the bruised area was marked with an ink circle, approximately 1 cm in diameter. From a total of 60 samples, only 40 samples were damaged using above technique, and the rest were maintained as non-bruised controls.

### 2.2. OCT image acquisition

The SD-OCT device (Telesto, Germany) was manufactured by Thorlabs, and its axial and lateral resolutions were set at $9.6 \times 10^{-6}$ m and $3.48 \times 10^{-6}$ m, respectively. The device was operated at $1300 \times 10^{-9}$ m, and got the moderate penetration depth and resolution. Because of both light absorption and scattering existing in fruit tissue, the light at a lower wavelength (e.g, $800 \times 10^{-9}$ m) can not penetrate the peel of the loquat, and the wavelength of 1300 × 10–9 m was compromised with penetration depth and light attenuation. The light beam was directly focused on the skin of the loquat through an integrated probe (LSM03, Germany). The back-scattered photons were also captured by the same probe with the reference path length and intensity adjusted according to the instructions for using the OCT device (Ruggeri et al., 2016). The X and Z direction included 941 and 1024 pixels respectively. Under the current axial and lateral resolution settings, and the field of view of image was $9.04 * 3.57 \times 10^{-3}$ m. In the following experiment, each estimation of the attenuation coefficient only required 400 A-scans and 250 points from the lateral direction, which was very small area and had a better ability to mitigate against disturbances caused by the curvature effect. The SD-OCT device can obtain the 3D dataset with the support of a galvanometer in a probe by acquiring depths scans at adjacent lateral positions. However, the attenuation coefficients in our study were estimated from sectional images, and only the 2D mode was used in following experiment.

There was no requirement for sample pretreatment, and the OCT raw images were acquired from both non-bruised and bruised samples after mechanical injury was applied. In addition, we also acquired the raw images of bruised samples at 12 h from the initial injury. We did not use bruised samples after more than 12 h because the skin color changed after that.

### 2.3. Image processing

Before quantitative estimation of the attenuation coefficients of the loquat sub-surface tissue, the task of image processing was to obtain the appropriate image from the OCT model (discussed in the following section). One OCT image contained upper and lower parts, the background and loquat target. The analysis object was the target part, so segmentation became an important preprocessing step. Unfortunately, most of the pixel grayscales at the background position were not equal to zero (ideal black) due to contamination by speckle noise. The threshold denoising approach was used for suppressing noise with an empirical threshold $\mu + 7 * \sigma$ in which the mean ($\mu_t$) and variance ($\sigma$) of grayscales of the top 20 rows in the background part of the OCT image were components of the threshold (Dong et al., 2013). Next, the edge between the target and background was detected by grayscale difference. Owing to the unsmooth nature of the detected edge, it was required that the loquat curvature be flattened. The unsmoothed edge was fitted by a polynomial curve, and points in the target area were shifted up and down, making all the points of edge lay on a horizontal line. Fig. 1 demonstrates the flattening process, where Fig. 1(b) is the version of Fig. 1(a) with the target curvature flattened. After flattening, we cropped pixels above the edge from the flattened image.

### 2.4. OCT model for attenuation coefficients

The relevant reports on fruit optical properties showed that loquat flesh was a type having weak scattering characteristics, and it existed both single and multiple scattering effects (Seifert et al., 2015; Hu et al., 2015; Lu et al., 2017). For weakly scattering media, comparative studies of the two effects have been carried out, with a conclusion that a single scattering model should be used for describing the scattering phenomenon. The OCT device only utilized the depth resolved information from the interference light by adjusting the coherence path/length