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EVALUATION OF CHILLING INJURY SYMPTOMS IN BANANAS USING OPTICAL IMAGING METHODS

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By

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Chairman: Rimfiel bin Janius, PhD

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The development of chilling injury symptoms in bananas (*Musa cavendishii*) at different ripening stages (2 to 5) was investigated using optical imaging methods. A total of 240 bananas with 60 bananas per each ripening stage ripened in a commercial ripening room were stored at 6 and 13 °C for two days to induce chilling injury symptoms. Images of the bananas were acquired using RGB and backscattering imaging at before storage (T0), during storage i.e. after 24 hours of storage with 90-minute time intervals (T1 to T5) and after storage i.e. exposure to ambient temperatures with the same time interval (T6 to T10), to monitor the appearance of chilling injury symptoms. The captured colour images were analyzed and valuable information from the images was extracted. Backscattering images were acquired using laser diodes emitting at 660 nm (visible light) and 785 nm (near-infrared light) wavelengths. The backscattering images were analyzed based on the changes of intensity in the radial direction providing a curve of intensity, also known as the backscattering profile. The backscattering parameter such as inflection point (*IP*), slope after inflection point (*SA*), full width at half maximum (*FWHM*) and saturation

radius (*RSAT*) were extracted from the backscattering profile to relate with the appearance of chilling injury symptoms.

The changes in all the optical parameters as a function of time were investigated. The changes in colour parameters were compared with visual assessment (*VA*) values while the changes in backscattering parameters were compared with pigment and water contents. Since *VA* is a non-destructive method, the measurement was conducted immediately after measurements using optical imaging methods (T0 to T10). The pigment and water contents measurements, due to their destructive methods, were carried out at the end of the experiment (T10). Significant ($P<0.05$) changes in all optical parameters, *VA*, pigments and water content subjected to temperature, ripening stage and treatment time were obtained. The analysis of the changes in colour parameters during treatment time provided the onset values of colour and time for development of chilling injury. The rate of reaction for the colour changes during the appearance of chilling injury symptoms were investigated using zeroth, first- and second-order model. Results revealed that hue (*H*) was the most suitable colour parameters to evaluate the development of chilling injury symptoms. The changes in *H* during development of chilling injury symptoms for each ripening stage can be predicted using zeroth-order model and first-order model for during and after storage, respectively. For during storage, kinetic model for each ripening stage provided lower uncertainty than irrespective ripening stages with $R\geq0.970$. For after storage, kinetic model irrespective ripening stages provided comparable results with each ripening stage with R varies from 0.936 to 0.979 for analysis with each ripening stage and $R=0.976$ for analysis irrespective ripening stages. Analysis of the changes in backscattering parameters revealed that backscattering imaging at 660 nm obtained lower uncertainty than 785 nm with misclassification error less than 7% for 660 nm and 20% for 785 nm. Results also revealed that *FWHM* and the combination of *IP*, *SA* and *FWHM* for 660 nm and 785 nm respectively were the most appropriate backscattering parameters for evaluating chilling injury. Hence, optical imaging methods i.e. RGB and backscattering imaging are potentially useful in the evaluation of chilling injury symptoms in bananas.

Abtsrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

PENILAIAN SIMPTOM-SIMPTOM KECEDERAAN DINGIN BUAH PISANG MENGGUNAKAN KAEDAH-KAEDAH PENGIMEJAN OPTIK

Oleh

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Pembentukan simptom kecederaan dingin pada buah pisang (*Musa cavendishii*) daripada tahap kematangan yang berbeza (2 hingga 5) diselidiki dengan menggunakan kaedah-kaedah pengimejan optik. Sejumlah 240 buah pisang, dengan 60 buah pisang daripada setiap tahap kematangan yang dibiarkan matang di bilik sejuk komersil, disimpan pada suhu 6 dan 13 °C selama 2 hari untuk mendorong kemunculan simptom kecederaan dingin. Imej-imej buah pisang tersebut dirakam menggunakan pengimejan RGB dan serakbalikan pada sebelum penyimpanan (T0), semasa penyimpanan iaitu selepas 24 jam dengan selang 90-minit (T1 hingga T5) dan selepas penyimpanan iaitu selepas pendedahan kepada suhu sekitar dengan tempoh selang yang sama (T6 hingga T10), untuk memerhati pembentukan simptom kecederaan dingin. Imej-imej yang dirakam dengan menggunakan pengimejan RGB dianalisa dan informasi yang bernilai daripada imej-imej tersebut diekstrak. Imej-imej serakbalikan dirakam menggunakan peranti diod laser yang memancarkan cahaya pada gelombang 660 (cahaya nampak) dan 785 nm

(cahaya hampir inframerah). Imej-imej serakbalikan tersebut dianalisa berdasarkan perubahan kecerahan dalam arah radial yang menghasilkan lengkung kecerahan, juga dikenali sebagai profail serakbalikan. Parameter-parameter serakbalikan iaitu titik infleksi (*IP*), kecerunan selepas titik infleksi (*SA*), lebar lengkap separa maksimum (*FWHM*) dan jejari saturasi (*RSAT*) kemudiannya diekstrak daripada profail serakbalikan untuk mengaitkannya dengan pembentukan simptom kecederaan dingin.

Perubahan semua parameter optik dengan masa diselidik. Perubahan parameter-parameter warna dibandingkan dengan taksiran visual (*VA*) manakala perubahan parameter-parameter serakbalikan dibandingkan dengan kandungan pigmen dan air. Memandangkan *VA* adalah kaedah tanpa musnah, pengukuran *VA* dijalankan sejurus selepas pengukuran menggunakan kaedah-kaedah pengimejan optik (T0 hingga T10). Bagi pigmen dan kandungan air, disebabkan kaedah pengukurannya adalah secara musnah, pengukuran hanya dijalankan pada akhir eksperimen (T10). Perubahan yang signifikan ($P<0.05$) didapati pada semua parameter-parameter optik, *VA*, kandungan pigmen dan air terhadap suhu, tahap kematangan dan masa rawatan. Analisis perubahan parameter-parameter warna berlandaskan masa menghasilkan nilai permulaan warna dan masa untuk pembentukan kecederaan dingin. Kadar perubahan tindakbalas semasa pembentukan simptom kecederaan dingin disiasat menggunakan model tertib sifar, pertama dan kedua. Keputusan menunjukkan bahawa klasifikasi warna (*H*) adalah parameter warna yang paling sesuai untuk menilai pembentukan simptom kecederaan dingin. Perubahan *H* semasa pembentukan simptom kecederaan dingin boleh diramal menggunakan model tertib sifar dan model tertib pertama, masing-masing untuk semasa dan selepas penyimpanan. Semasa penyimpanan, model kinetik bagi setiap tahap kematangan menghasilkan ketidaktentuan yang lebih rendah berbanding model kinetik yg mengabaikan tahap kematangan dengan R≥0.970. Selepas penyimpanan, model kinetik yang mengabaikan tahap kematangan menghasilkan keputusan yang setara dengan model kinetik bagi setiap tahap kematangan dengan R bernilai dari 0.936 ke 0.979 untuk analisis bagi setiap tahap kematangan dan R=0.976 untuk analisis yang mengabaikan tahap kematangan. Analisa perubahan parameter-parameter serakbalikan mendedahkan bahawa pengimejan serakbalikan pada 660 nm menghasilkan ketidaktentuan yang lebih rendah berbanding 785 nm dengan kesilapan pengkelasan kurang dari 7% untuk 660 nm dan 20% untuk 785 nm. Keputusan juga mendedahkan bahawa *FWHM* dan kombinasi *IP*, *SA* dan *FWHM*, masing-masing untuk 660 dan 785 nm adalah parameter serakbalikan yang paling sesuai untuk menilai kecederaan dingin. Maka, kaedah-kaedah pengimejan optik iaitu pengimejan RGB dan serakbalikan berpotensi sangat berguna untuk menilai simptom kecederaan dingin buah pisang.

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I certify that a Thesis Examination Committee has met on 23rd November 2012 to conduct the final examination of Norhashila binti Hashim on her thesis entitled "Evaluation of Chilling Injury Symptoms in Bananas Using Optical Imaging Methods" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U. (A) 106] 15 March 1998. The Committee recommends that the student be awarded the Doctor of Philosophy.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any degree at Universiti Putra Malaysia or at any other institutions.

NORHASHILA BINTI HASHIM

Date: 23 November 2012

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CHAPTER 1

INTRODUCTION

1.1 General Overview

The banana is one of the tropical fruits that establishes well in the market and is commercialized all over the world. The fruit is nutritious, easy-to-eat and can be processed into a number of food products such as ice-cream, cake, baby food, bread and many more. The production and demand for bananas increased yearly, making it one of the most important foods in the world and ranked fourth behind rice, wheat and maize in the world food ranking (FNCA, 2011). According to the Food and Agricultural Organization of the United Nations (FAO) database, the world total production of bananas increased steadily between year 2000 and 2009 (Figure 1.1).

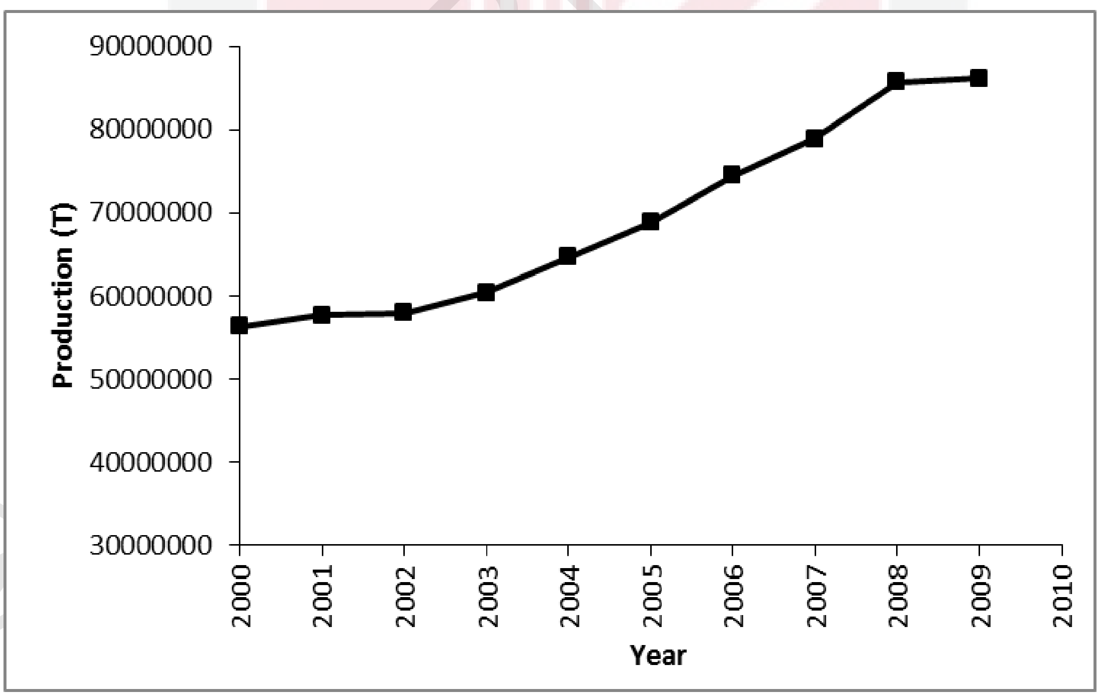

Figure 1.1: Total world production of bananas by year.
(Source: FAO, 2012)

A sharp rise can be seen beginning from 2002 to 2008 which accounted for 70.1 million tonnes average per year. The rapid growth was reported to be due to the expansion of planted areas in Ecuador and the Philippines in the late nineties (FAO, 2012). In 2009, the world total production for bananas has reached a maximum value of 86 million tonnes. This positive trend shows the bright future of the banana industry in the world market.

Presently, quality is the main concern in human daily life. As the demand for higher quality food and produce increases, the ability to manufacture high quality produce consistently becomes the basis for success in the highly competitive food industry (Gunasekaran, 1996). Li and Jia (2008) reported that between 20-30% of fruits were lost due to poor handling and storage after harvest. The massive loss during postharvest handling may run into billions of dollars worldwide (Bapat et al., 2010). Thus, many attempts have been made to maintain and improve the quality of the produce. One of the most practical methods is storing the produce at low temperature to preserve its quality. However, when fresh produce is exposed to a temperature below its tolerance threshold, chilling injury symptoms might appear which would strongly affect its quality and marketability.

Chilling injury was defined as the physiological damage that occurred at temperatures above freezing (0 °C) and generally below 10°C (Wills et al., 1998). The banana is one of the chill-sensitive produce which showed an initiation of chilling injury when subjected to a temperature below 10°C (Murata, 1969; Abd El-Wahab and Nawwar, 1977; Broughton and Wu, 1979; Nguyen et al., 2003). The most common symptoms of chilling injury were dark skin areas, a scald-like discoloration, and pitting or sunken lesions. Such chilling injury symptoms were due to the disruption of the physiological and physico-chemical changes during the ripening process. For bananas, the most common symptoms of chilling injury was browning or discolouration in which the colour of the bananas changed from green or yellow to brown and eventually completely black when the symptom became severe (Murata, 1969; Abd El-Wahab and Nawwar, 1977; Broughton and Wu, 1979; Nguyen et al., 2003). Chill-injured fruits also underwent a change in texture due to the breakdown of the membrane and cell walls. Ratule et al. (2006) reported that the exposure of bananas to chilling temperature for a long time would cause hardening of the flesh. Such disorder would decrease the quality of the fruit and repel consumer acceptance.

There have been tremendous developments in non-destructive techniques for the detection of food and fruit quality. Near-infrared spectroscopy (NIRS) is an optical technique that has been widely used by researchers in recent years. Slaughter et al. (1998) found that near-infrared spectroscopy could predict the fructose, glucose, soluble solids content (SSC) and dry weight of kiwifruit with coefficient of correlation R being greater than 0.95. The method also showed significant results in the measurement of internal quality of tomatoes (He et al., 2005), the evaluation of SSC in watermelons (Tian et al., 2007) and the evaluation of SSC and firmness in pears (Liu et al., 2008). Zude et al. (2006) reported that although a correlation between changes in chlorophyll and fruit flesh firmness by the Magness-Taylor test exists, robust calibration of fruit flesh firmness using this correlation is hardly possible. The correlation between firmness using the NIRS technique and the conventional method was low when validating the calibration model with an independent test-set (McGlone and Kawano, 1998; Lu et al., 2000; Zude et al., 2006; Noh and Lu, 2007).

Computer vision is one of the advanced non-destructive techniques that receive tremendous attention due to its effectiveness and accuracy. Computer vision system together with a colour camera was used to evaluate colour and firmness of tomatoes (Edan et al., 1997), measure colour changes in mangoes during fruit development (Kang et al., 2008) and detect external damage of olives (Requelme, 2008). The technique could quantitatively characterize complex sizes, shapes, colours and textural properties of foods while eliminating the subjectivity of manual inspections (Du and Sun, 2004). Du and Sun (2004) further stated that the technique offers flexibility in application and can be reasonable substitute for human visual decision-making process.

Lu (2004) combined vision system and spectral readings using a laser diode light source to develop backscattering imaging which produce backscattered images. The backscattered images were analyzed and the interaction between the fruit tissue and the apparent intensity and path length of light from the incident point until total attenuation was used to obtain information on the structural and chemical characteristics of the fruit. The technique provided promising result in the detection of many fruit qualities such as apples (Peng and Lu, 2006; Qing et al., 2007; Qin and Lu, 2008; Huang and Lu, 2010), peaches (Lu and Peng, 2006), pear, plums, tomatoes, zucchini, cucumber (Qin and Lu, 2008) and kiwifruit (Qin and Lu, 2008; Baranyai and Zude, 2009).

1.2 Problem Statement

Although numerous studies on fruit quality using advanced non-destructive techniques have been conducted on fresh produce in general and bananas in particular, no information is available on measuring the development of chilling injury symptoms in bananas. Most of the reports concerned quality changes during the ripening process. Since the banana is one of the most important yet sensitive tropical fruits, detection of chilling injury is vital to ensure that only high quality produce reaches the consumers.

Chilling injury is a time and temperature dependent problem, being strongly dependent on the duration and temperature of chilling treatment as well as the period of post-chilling storage. Detection and diagnosis of chilling injury are often difficult as produce often look sound when removed from the chill environment and symptoms may only develop when the produce is placed at higher temperatures. They may appear almost immediately or after several days (Skog, 1998). Symptoms also may be invisible externally and sometimes a very slight browning discoloration, which is actually the first appearance, has a tendency to be misclassified as normal by untrained inspectors (Skog, 1998; ElMasry et al., 2009). Such delayed appearance of chilling injury symptoms creates problems in transportation, storage, and marketing (Murata, 1969) leading to economic losses (Liu, et al., 2006).

The extent of chilling injury damage is also hard to quantify and is commonly estimated using visual inspections and destructive methods. The visual inspection method, although rapid and easy, is known to have many disadvantages. It is a subjective method that depends very much on the competence of the human worker who is influenced by many factors such as physique and emotion causing reliability problems. As reported by Bollen et al. (1993), the ability of a human being to perceive visual images depends on the cognitive factors of the person whose visual acuity decreases with age. Colour perceived by human is strongly influenced by the intensity of the light received by the eyes. An older worker may require about twice the brightness level of the younger worker for an equivalent result. This situation could lead to high variability and inaccuracy resulting in non-uniform quality of the produce. Also, the destructive method damages the produce, hence does not have repeatability, rendering them quite unsuitable for the market. Therefore, it is vital that rapid, sensitive and non-destructive chilling injury evaluation be performed to ensure fruit quality and marketability.

1.3 Objectives

The aim of this study was to evaluate chilling injury symptoms by means of non-invasive optical imaging methods i.e. RGB and backscattering imaging. The specific objectives of this study were:

a) To investigate the changes in colour and backscattering parameters during the development of chilling injury in bananas
b) To determine the best colour and backscattering parameters that could describe chilling injury in bananas.
c) To determine the onset of skin colour and time of chilling injury in bananas.
d) To determine the rate of skin colour change in bananas during chilling injury development.

1.4 Scope and Limitation of the Study

The bananas studied were the *Musa cavendishii* cultivar at ripening stages 2 to 5, targeted for the export market.

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