CAFEi2012-219

EVALUATION OF CHILLING INJURY IN BANANAS USING BACKSCATTERED LASER IMAGING

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ABSTRACT

The change in backscattering parameters during the appearance of chilling injury in bananas was investigated. Bananas were stored at a chilling temperature for two days and the degrees of the chilling injuries that appeared were measured before, during and after storage using backscattering imaging and visual assessment. Laser lights at 660 nm and 785 nm wavelengths were shot consecutively onto the samples in a dark room and a camera was used to capture the backscattered lights. Plots of light intensity of the images against pixel count provided a backscattering profile which contained useful information on backscattering parameters such as inflection point (IP), slope after inflection point (SA), full width half maximum (FWHM) and saturation radius (RSAT). Results indicated that there were significant changes in backscattering parameters as chilling injury developed.

Keywords: *banana, chilling injury, backscattering, fruit quality, imaging.*

INTRODUCTION

As a chill-sensitive produce, bananas are susceptible to chilling injury when exposed to chilling temperatures. Since the banana is the 4th most important food behind rice, wheat and maize in the world food ranking [1], chilling injury becomes a major problem in marketability in the banana industry. The shipment of the fruits from the producer countries to the importing countries requires the fruits to be stored in cold storage to ensure that they will be in a good condition upon arriving at their destinations. Bananas were reported to exhibit chilling injury symptoms when exposed to a temperature below 10 °C [2-5]. As a consequence, the injured fruit showed discoloration or browning symptom, in which the green or yellow colour of the skin changed to brown and completely black, depending on the severity of the injury.

The appearance and severity of the injury was strongly influenced by temperature, time and ripening stage [2, 6-7]. The severity increases and the appearance deteriorate faster with lower storage temperatures and longer exposure to chilling temperature. The symptoms become clearer after the fruit is removed from cold storage and the appearance of the symptoms will be exhibited immediately or takes several days in which case there is high possibility that the infected fruit could escape detection during the sorting process which in turn will affect its marketability. In addition, the conventional method of chilling injury detection used i.e. visual assessment (VA) is easily exposed to human error due to the dependency of the technique on human visual skill. Thus, a more advance technology which is non-destructive, inexpensive, faster and accurate is needed to solve the problems.

Backscattering imaging is one of the advance methods that is being considered for quality determination of agricultural produce in recent years. The method physically applies a theory of interaction between light and agricultural materials. As reported by [8], when light hits a tissue, 4% of the light will be reflected back to the atmosphere while the rest will penetrate and being absorbed, transmitted or scattered back (diffuse reflectance) to the incident point. The interaction of light during penetration in the fruit tissues carries much useful information about the structure of the material which later could be used to measure the quality of the produce.

Image acquisition using backscattering imaging provides backscattering images that consist of a circular illumination spot in which the intensity of the illumination decreases radially outwards. [9] reported that the outer part (low level intensity) of the illumination spot in the image showed high correlation with the change in the total number of pixel, thus carrying the most information about the characteristics of the tissue. [10] found that a histogram of backscattering intensities is highly correlated with soluble solids content (SSC) of apples. Backscattering parameters obtained from the curve fitting of backscattering intensities such as inflection point (IP), full width at half maximum (FWHM), slope and etc. correlated well with the textural properties of peach [11], apples [12-13], ripeness stages of tomatoes [14] and, more recently, the moisture content of bell pepper [15]. Since there were promising results obtained in the application of backscattering imaging in agricultural produce, the effect of chilling injury in bananas on the backscattering parameters was studied in order to evaluate the ability of the method in replacing the conventional method of VA.

MATERIALS AND METHODS

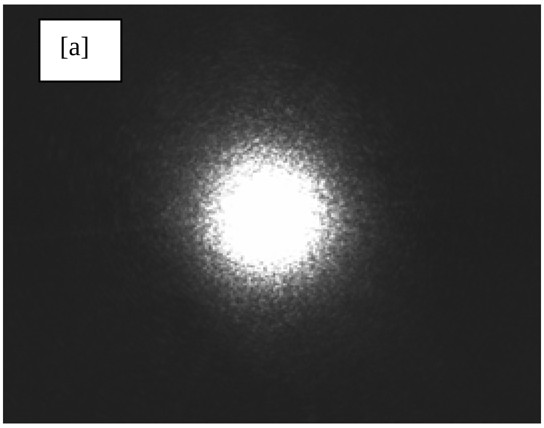
Fruit samples

Musa cavendishii bananas from ripening stages two, three, four and five were obtained from a commercial banana ripening facility (FruchtExpress Import Export GmbH, Germany). Samples were evenly divided into two groups, i.e., the chill-treated samples which were stored at a chilling temperature of 6 °C and the control samples which were stored at 13 °C. Both groups were stored for two days before taken out and exposed to ambient temperatures. Data collection was carried out at before storage (day 1), during storage (day 3) and after storage (day 4) by using backscattering imaging. Data obtained by the visual assessment method were taken as the reference.

Image acquisition

Backscattering images of bananas were recorded using an in-house-developed laser-induced backscattering imaging system in the Department of Horticultural Engineering, Leibniz Institute for Agricultural Engineering, Potsdam-Bornim (ATB), Germany. Each banana was placed under a CCD camera (JVC KY-F50E) with zoom lens (F2.5 and focal lengths of 18-108 mm). Laser diode of 1 mm beam size emitting at 660 and 785 nm with 45 mW maximal power was used as a light source. Backscattering images of sizes 720 x 576 pixels were acquired in a dark-room in order to obtain a good signal-to-noise ratio. A total of six images consisting of 3 images per side of a banana were taken to obtain the average value of the backscattering for each fruit. The Lambertian cosine law was applied to adjust the intensity values of the surface captured by the CCD camera. An in-house-developed software was installed in the computer to assist in the image acquisition process.

The backscattering images were identified by the brightness of the light (Fig. 1a) which decreased radially outwards as the distance from the illumination point increased, providing a symmetric backscattering profile (Fig. 1b). From the backscattering profile, the backscattering parameters, i.e., the inflection point (IP), the slope after inflection point (SA), the full-width-half-maximum (FWHM) and the saturation radius (RSAT) were obtained. The IP was defined as the minimal value of the first derivative of the profile. The FWHM was given by the distance between two points on the curve at which the profile reached half its maximum value. The RSAT is the distance between the incident point and the IP of the backscattering profile. Values of the backscattering parameters obtained from the analysis were then transferred as text files to Matlab or MSExcel for statistical analysis.



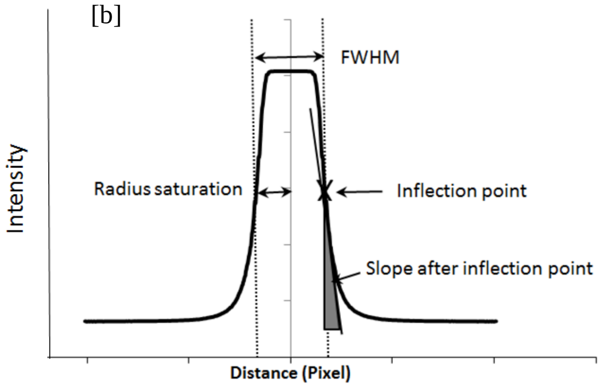


Fig. 1: (a) A raw backscattering image acquired using a 785 nm laser and (b) the backscattering profile and parameters of the raw image.

Visual assessment

The visual assessment method using a browning scale as described by [5] was performed immediately after the backscattering image acquisition. The browning scale was rated as follows: 1 = no chilling injury symptoms; 2 = mild chilling injury symptoms in which injury can be found in between the epidermal tissues; 3 = moderate chilling injury symptoms in which brown patches begin to become visible, larger and darker; 4 = severe chilling injury symptoms in which the brown patches are clearly visible and are larger and darker than at scale 3; 5 = very severe chilling injury symptoms in which the patches are relatively large on the surface.

Data analysis

Descriptive statistics and analysis of variance (ANOVA) were carried on the data obtained using Matlab (Math Work Inc., USA) and SAS statistical software.

RESULTS AND DISCUSSIONS

Evaluation by visual assessment data

All the bananas stored at the control temperature (13 °C) were not affected by chilling injury after removal from storage and being exposed to ambient temperature; all had a VA value of 1. The fruits ripened normally and developed a golden yellow colour due to the breakdown of chlorophyll (green pigment) thereby unmasking the carotenoids (yellow-red pigment). The pulp became tender and soft thus making the fruit edible

e-ISBN 978-967-960-322-4

fresh which, as reported by [16], was due to the depolymerization and solubilization of pectins (carbohydarate molecules in the cell walls). This indicated that the temperature of 13 °C is suitable for storage with no initiation of chilling injury to the samples. The present study together with the findings of [17] and [18] confirmed that the slowing down of normal ripening of bananas and the prolonging of the fruit shelf life can be achieved by storing at 13 °C.

In contrast, the chill-treated bananas (6 °C storage) did not ripen after being exposed to ambient temperature but exhibited discoloration in which the colour of the skin changed from green or yellow to brown or black, a phenomenon which can be attributed to the accumulation of phenolic substances. This means that the ripening process of the chill-treated samples was halted and, due to the chilling injury, phenolic substances were being oxidized. The pulp also tended to be harder indicating that the process of disassembly of the cell wall and conversion of carbohydrates to sugar did not happen thus making the fruit to become off-flavours. Different parts of these observations were observed also by [18], [5] and [19]. Values of visual assessment for the chill-treated bananas at different ripening stages are as illustrated in Fig. 2.

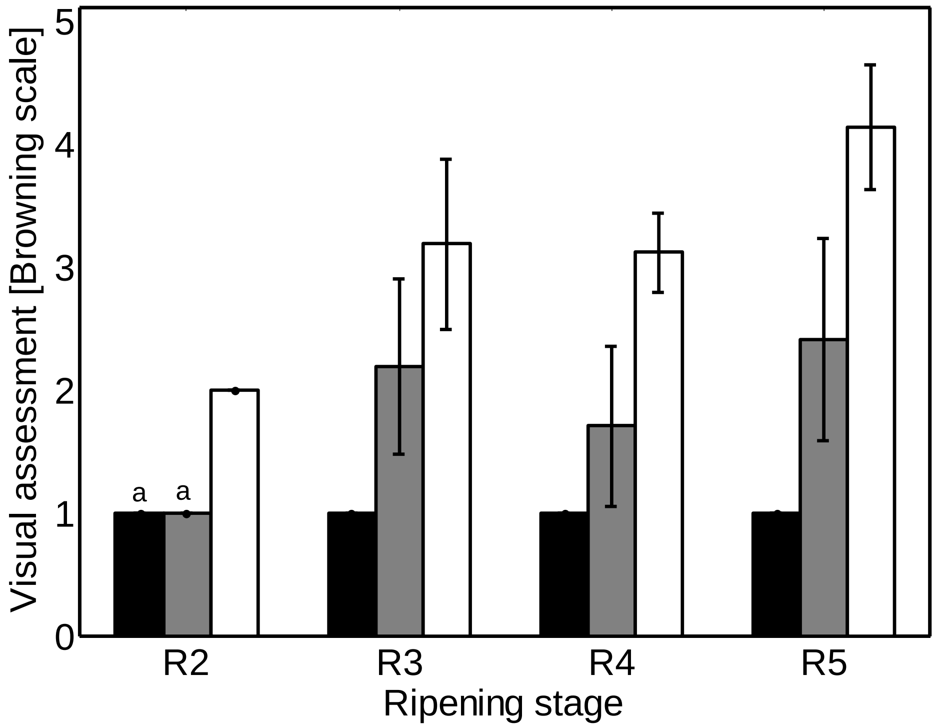


Fig. 2: Visual assessment values of bananas at different ripening stages stored at 6 °C (■: Before storage, ■: During storage, □: After storage). Bars represent mean ± standard deviation. The letter *a* indicates values that are not significantly different (p>0.05)

The mean values for bananas stored at 6 °C increased from before to after storage indicating there was a change in the skin colour of the fruits upon exposure to chilling temperature. Ripening stage R2 obtained the lowest mean values compared to the more advanced ripening stages either during or after storage denoting that the skin colour were less affected by the chilling temperature. The degree of browning had a value of 2 which indicated that the browning symptom was at a mild stage. Although the browning was very slight, the injury was, nevertheless, severe. The bananas failed to ripen normally due to the failure of the fruit to produce ethylene. As a result, although the injured fruits maintained green they became very hard and were not fit for human consumption.

For the more advanced ripening stages (R3, R4 and R5), the mean values changed from 1 (before storage) to a maximum of 4 (after storage) demonstrating that the browning symptom had developed and the severity increased as the treatment time progressed. The metabolism change was the same as in the R2 samples. However, the colour of the fruits of the more advanced stages before being stored in chilling temperature had already turned yellow and the pulp already soft due to the ripening process. Since the degreening process of the skin had already taken place even before storage, the symptoms of chilling injury could be easily detected from the colour contrast between the browning and the light yellow colour of the skin. In addition, the disassembly of the cell walls and conversion of carbohydrates to sugar which contributed to the softening of the ripe fruit had exposed the fruits to diseases or fungal infections which could facilitate mechanical injury and decay. As a result, as the stage of ripening advanced, the severity of injury increased in tandem with the appearance of browning. This finding was in agreement with [2] and [20] who reported that the appearance of chilling injury symptoms becomes severe as the maturity level increases. The degree of browning also increased when the samples were exposed to ambient temperature. Thus, it can be inferred that time, temperature and maturity stage had strong influences in the appearance of chilling injury.

Evaluation by backscattering parameters

The values of backscattering parameters measured using 660 nm laser for chill-treated bananas at before, during and after 6 ^{o}C storage are shown in Fig. 3.

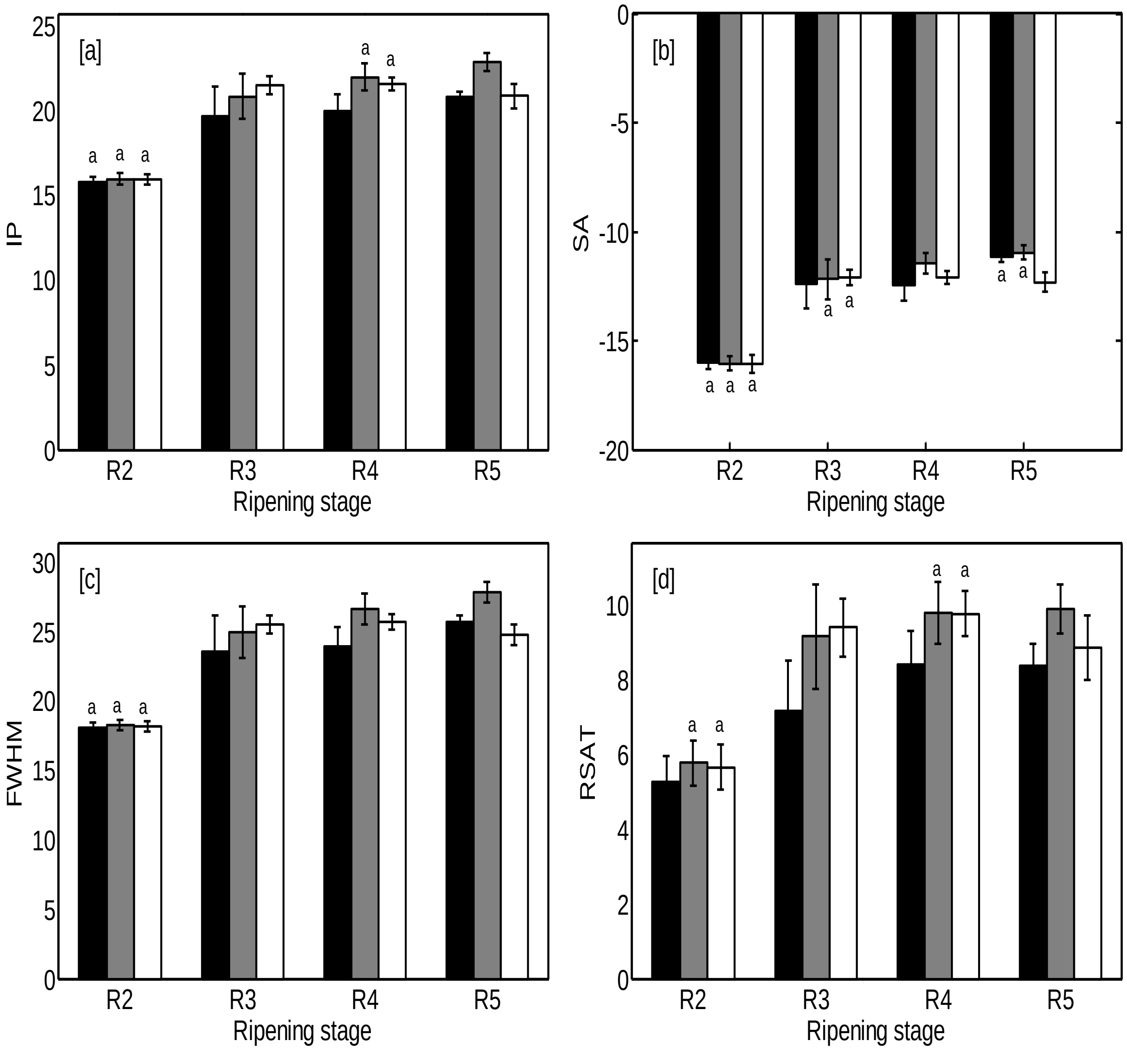


Fig. 3: Values of various backscattering parameters at 660 nm of bananas at different ripening stages stored at 6 °C (■: Before storage, ■: During storage, □: After storage). Bars represent mean ± standard deviation. The letter *a* indicates values that are not significantly different ($p>0.05$)

Before storage, the values of the backscattering parameters for 660 nm increased as ripening stages increased reflecting the increase in the backscattering areas. Since the different ripening stages had different skin colour, this phenomenon could be explained to be due to the interaction between the wavelength and the chlorophyll which absorbed light at this wavelength. As the fruit ripened and maturity level increased, the chlorophyll pigment disappeared and carotenoids pigment appeared reflecting the degreening process, in which the colour changed from green to yellow. As a result, very little or no absorption of the light by chlorophyll took place and the photons were backscattered resulting in the observed increase of the backscattering area. Parallel to this observation is that of [15] in which less scattering and a decrease in the total pixel number of green bell peppers was reported and was explained to be due to the chlorophyll pigments absorbing light at approximately 670 nm bandpass.

When stored in chilling temperature, the fruits resulted in lower levels of chlorophyll than did the control samples. This was due to the severe damage in the chloroplast membranes which stored the chlorophyll pigment and to the acceleration of pigments degradation during chilling injury. The lesser amount of chlorophyll present to absorb the incident light resulted in an increased backscattering area whose profile was described by the backscattering parameters IP, SA, FWHM and RSAT. However, SA did not show any significant difference in changes from during to after storage for all ripening stages. Hence, SA will not be discussed any further.

For R2 and R3, the values of IP, FWHM and RSAT increased from during storage to after storage. For R4, these parameters did not change significantly in values and for R5 they decreased. However, each of these parameters had after-storage values that were about the same for R3, R4 and R5 and this could be due to the fruits being already severely injured that no chlorophyll was present anymore to effect any change in readings. Results for R2 were different from the others in that the parameters were essentially the same for before, during and after storage indicating that the chlorophyll content for this ripening stage was not affected by the chilling temperature.

While 660 nm is sensitive to pigment content, laser light at near-infrared wavelength (770-2500 nm) is sensitive to textural properties of the fruits [21]. The values of the collected backscattering parameters are as presented in Fig. 4. The IP and FWHM for 785 nm of R2, R3 and R4 decreased from before to after storage. This decrease could be related to the change in the textural properties of the bananas. Exposure of bananas to chilling temperature resulted in a decrease in turgor pressure of the fruits which in turn enhanced water losses. [22] explained that as chilling injury develops, water losses become greater due to cellular breakdown, deterioration of membrane integrity as well as loss of epicuticular wax which is important in controlling water exchange in the skin. Thus, the intercellular spaces become larger and filled with air promoting direct reflection or transmission of penetrated light instead of backscattering it. As a result, the backscattering area decreased; hence the lower values of during-storage IP and FWHM for the bananas. The injury became severe after storage and the backscattering area and backscattering parameter values decreased further.

After being exposed to chilling temperature, the bananas at R2, R3 and R4 became harder due to severe infection of chilling injury and gave about similar response to the penetrating laser light as reflected in the approximately same values of IP and FWHM (Fig. 4a and Fig. 4c). The R5 values for before and after storage were not significantly different. This could be due to the lesser solid component of the fruits available at R5 to scatter the penetrating light. The parameters SA and RSAT showed irregular trends which mean that SA and RSAT were not significantly influenced by the change in the texture of the bananas as chilling injury developed. Nevertheless, there were significant effects on the backscattering parameters due to chilling indicating the potential of employing backscattering imaging in the detection of chilling injury in bananas.

e-ISBN 978-967-960-322-4

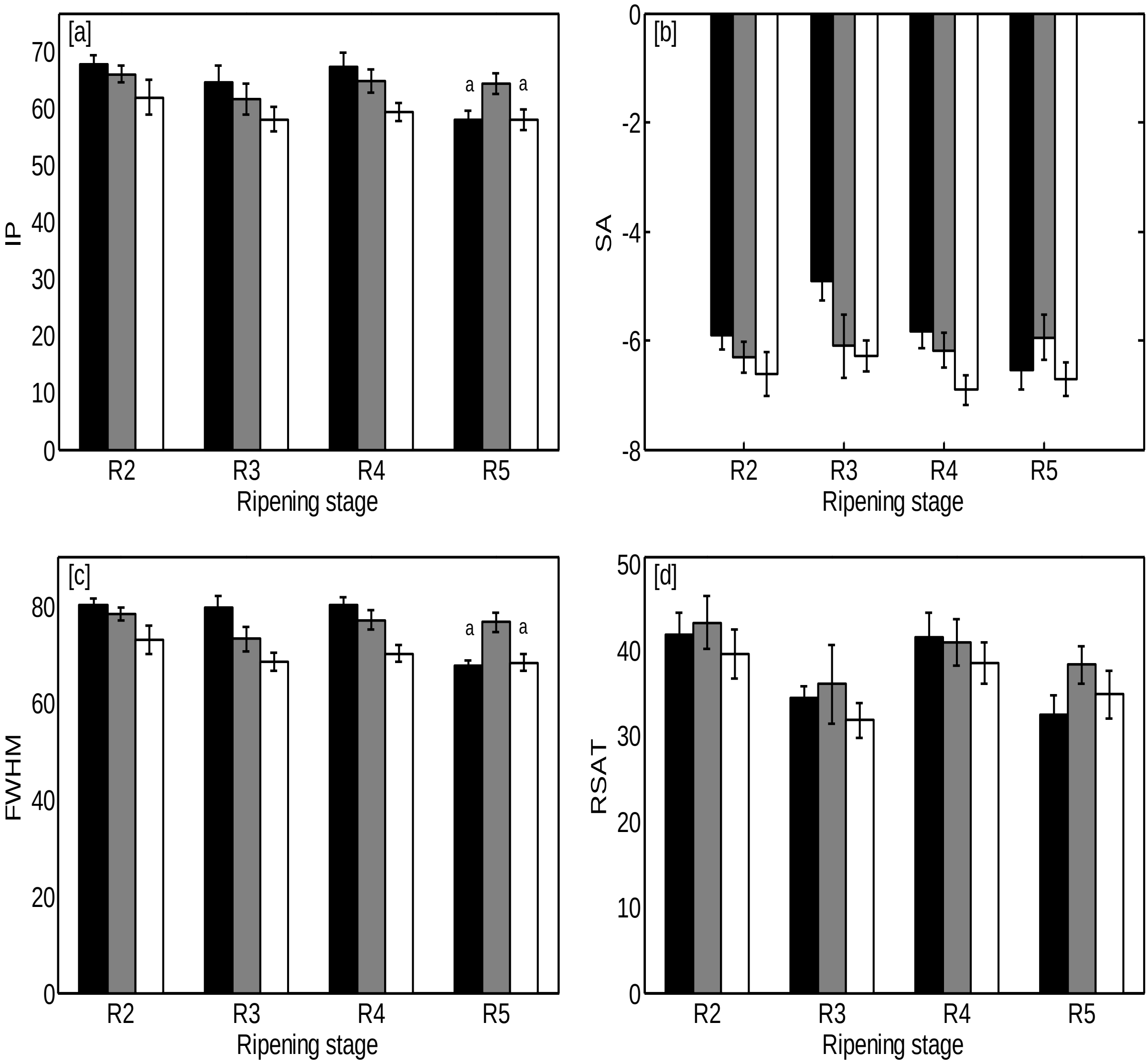


Fig. 4: Values of various backscattering parameters at 785 nm of bananas at different ripening stages stored at 6 °C (■: Before storage, ■: During storage, □: After storage). Bars represent mean ± standard deviation. The letter *a* indicates values that are not significantly different (p>0.05)

CONCLUSIONS

Laser-induced backscattering imaging using 660 and 785 nm wavelength lights have shown to be a good candidate for non-destructive detection of CI in bananas. The IP and FWHM parameters obtained from the backscattering profile showed significant changes from before to after storage. This means that the method can detect the change in colour and texture of bananas as chilling injury develops.

ACKNOWLEDGEMENT

The authors are grateful for the financial support received from the Ministry of Higher Education Malaysia, Universiti Putra Malaysia and Leibniz Institute for Agricultural Engineering Potsdam-Bornim (ATB) for this project.

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