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CHANGES IN SKIN COLOUR AND OTHER RELATED QUALITY CHARACTERISTICS OF 'B10' CARAMBOLA (AVERRHOA CARAMBOLA L.) AT DIFFERENT STAGES OF MATURITY

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By
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Thesis Submitted in Fulfilment of the Requirements for the Degree of Master of Agricultural Science in the Faculty of Agriculture
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CHANGES IN SKIN COLOUR AND OTHER RELATED QUALITY CHARACTERISTICS OF 'B10' CARAMBOLA *(Averrhoa carambola* L.) AT DIFFERENT STAGES OF MATURITY

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Chairman : Siti Hajar Ahmad, Ph.D

Faculty : Agriculture

A study was conducted on changes in skin colour and other related quality characteristics of 'B10' carambola *(Averrhoa carambola* L.) at different stages of maturity. In Experiment I the carambola were harvested at different stages of maturity beginning from 57 to 93 days after fruit-set with three day intervals between each harvest. Skin colour (L*, a*, b*, C* and h^O values), chlorophyll and carotenoid contents of the fruit were determined at each stage of maturity. In Experiment II other related quality characteristics such as flesh firmness, soluble solids concentration, sugar (sucrose, fructose, and glucose), ascorbic acid and organic acid (malic, citric and oxalic) contents at nine colour indices (as had been categorised in Experiment I) were also determined. Respiration rate and ethylene production of carambola were determined to study the climacteric nature of the fruit. Each of the experiment was conducted using a completely randomised design and the data obtained were analysed using analysis of variance and Duncan Multiple Range Test.

Simple regression model was used to determine relationship between maturity stages and skin colour (Experiment I) and relationship between colour indices and other related quality characteristics (Experiment II). Correlations among parameters in each experiment were obtained by means of Pearson's correlation matrix.

In Experiment I, a linear increase in a* and b* values indicated that skin colour of carambola had significantly lost its greenness and was turning yellow as fruit maturity increased. The linear decrease in h^O and linear increase in a*/b* values indicated that carambola skin colour had changed from green to yellow and yellowish orange. L* value increased linearly with increasing maturity, indicating that fruit skin colour became lighter as maturity increased. C* showed a linear increase as fruit matured, indicating that skin colour of the fruit had become intense yellow and yellowish orange. The h^O was the most accurate colour measurement predicted by maturity stages giving the highest coefficient of determination (r^2= -0.95) compared to other parameters. As such, the maturity stages of carambola were categorised into nine colour indices based on h^O value. The results showed that carotenoid pigment was present even before chlorophyll was degraded and it continued to increase with increasing fruit maturity. However, total chlorophyll pigment decreased with increasing fruit maturity. Chlorophyll and carotenoid pigment contents were significantly correlated with each of the parameters used to measure skin colour, indicating that chlorophyll and carotenoid pigments contributed to the skin colour changes of the carambola.

In Experiment II, there was a significant linear increase in soluble solids concentration as fruit matured. Significant linear increases were also found in fructose, glucose, sucrose and ascorbic acid contents with increasing fruit maturity. However, flesh firmness, malic and oxalic, except citric acid contents showed significant linear decrease with increasing fruit maturity. Soluble solids concentration, ascorbic acid, fructose, glucose and sucrose contents showed negative correlations with flesh firmness. Oxalic, malic and citric acid contents were positively correlated with flesh firmness. Significant linear regression between colour indices and other related quality characteristics indicated that quality characteristics of carambola could be estimated by colour indices. The respiration and ethylene production patterns classified the carambola as a non-climacteric fruit. Therefore the carambola should be allowed to remain attached to the tree until the fruit has attained the appropriate quality characteristic levels before being harvested.

Abstrak Tesis Yang Dikemukakan Kepada Senat Universiti Putra Malaysia
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PERUBAHAN WARNA BUAH BELIMBING 'B10' (*Averrhoa carambola* L.)
DAN CIRI-CIRI KUALITI YANG BERKAITAN PADA PELBAGAI
PERINGKAT KEMATANGAN

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Kajian telah dijalankan ke atas perubahan warna buah belimbing 'B10' (*Averrhoa carambola* L.) dan ciri-ciri kualiti yang berkaitan pada pelbagai peringkat kematangan. Pada kajian I, buah belimbing dituai dengan selang masa 3 hari pada pelbagai peringkat kematangan mulai daripada 57 sehingga 93 hari setelah berputik. Perubahan warna (nilai L*,a*,b*,C* dan h^{O}), kandungan pigmen karotin dan klorofil buah belimbing ditentukan pada setiap selang masa tersebut. Pada kajian II, ciri-ciri kualiti seperti kekerasan buah, jumlah pepejal terlarut, kandungan gula (fruktosa, glukosa dan fructosa), asid askorbik, asid-asid organik (oskalik, sitrik dan malik) pada kesembilan indek warna yang telah diperolehi daripada kajian I telah ditentukan. Kadar respirasi dan pengeluaran etilena juga ditentukan untuk mengkaji jenis respirasi buah belimbing. Setiap kajian dijalankan menggunakan rekabentuk rawak lengkap dan data yang diperolehi dianalisis menggunakan kaedah analisis varian dan Duncan

Multiple Range Test. Kaedah regresi digunakan untuk mengkaji hubungan antara peringkat kematangan dengan warna buah (kajian I) and hubungan antara indeks warna dengan ciri-ciri kualiti (kajian II). Kaedah korelasi matriks Pearson's dijalankan untuk mengkaji kaitan antara parameter pada setiap kajian.

Pada kajian I didapati bahawa nilai a* dan b* meningkat secara linear menunjukan bahawa warna buah kehilangan kehijauannya dan warna kuning kelihatan dengan bertambahnya kematangan buah. Nilai h^O menurun secara linear dan nilai a*/b* meningkat secara linear menunjukkan perubahan warna buah belimbing dari hijau ke kuning dan oren kekuningan. Nilai L* meningkat secara linear menunjukkan bahawa warna buah menjadi lebih cerah dan nilai C* meningkat secara linear menunjukkan bahawa warna buah tersebut semakin pekat dengan bertambahnya kematangan buah. Nilai h^O adalah paling sesuai dianggarkan (r^2 = -0.95) pada setiap peringkat kematangan berbanding dengan parameter lainnya. Berasaskan kepada nilai h^O peringkat kematangan buah belimbing dapat di bahagikan kepada sembilan indek warna. Hasil kajian mendapati kandungan pigmen karotin telah sedia ada sebelum pigment klorofil berkurangan dan kandungan karotin terus meningkat dengan bertambahanya kematangan buah. Kandungan pigmen klorofil berkurangan dengan bertambahnya kematangan buah. Adanya korelasi bererti antara kandungan pigmen klorofil dan karotin dengan parameter warna (L*, a*, b*, C* dan h^O) menunjukkan bahawa perubahan kandungan pigmen klorofil dan karotin menyebabkan perubahan warna buah belimbing.

Pada kajian II, kandungan jumlah pepejal terlarut meningkat secara linear dengan bertambahnya kematangan buah. Peningkatan secara linear juga didapati pada kandungan fruktosa, gluckosa, sukrosa dan asid askorbik dengan bertambahnya kematangan buah. Tetapi kekerasan buah, kandungan asid malik dan oksalik menunjukkan penurunan secara linear dengan bertambahnya kematangan buah. Asid sitrik tidak menunjukkan peningkatan secara linear dengan bertambahnya kematangan buah. Kandungan jumlah pepejal terlarut, fruktosa, glukosa, fructosa dan asid askorbik berkorelasi negatif dengan kekerasan buah. Manakala asid oksalik dan malik berkorelasi positif dengan kekerasan buah. Kandungan pepejal terlarut berkorelasi positif dengan kandungan fruktosa, glukosa, fruktosa dan asid askorbik dan berkorelasi negatif dengan asid oksalik, sitrik dan malik. Persamaan regresi linear yang bererti antara indeks warna dengan ciri-ciri kualiti kecuali asid sitrik menunjukkan bahawa ciri-ciri kualiti buah belimbing 'B10' dapat dianggarkan menggunakan indeks warna. Kadar respirasi dan pengeluaran etilena menunjukan bahawa buah belimbing 'B10' adalah buah bukan klimakterik. Oleh yang demikian buah belimbing harus dibiarkan di pokok sehingga mencapai kualiti yang sesuai sebelum dituai.

CHAPTER I

INTRODUCTION

Carambola (*Averrhoa carambola* L.) is also called the starfruit or star apple because it has five

with thin, smooth and glossy skin and a sweet-watery pulp, tasting like a cross between an apricot and a passion fruit (Macomboy, 1982). The fruit matures after 60-70 days of development, producing fruit that weigh between 80 and more than 200g (Campbell et al., 1989) and varying in size between 80-250 mm in length, and 50-100 mm in width (Watson et al., 1988). Fruit colour varies from green to yellowish orange depending on the cultivar and stage of maturity.

Carambola is one of the most important fruits for export. The total export of carambola has not increased since 1990. Total export was 11 metric tons in 1990 compared to 8.9 metric tons in 1995. Although total export has not increased, the value of carambola exports has increased from RM22.5 million in 1990 to RM23.60 million in 1995. The percentage of carambola exported (12%) was even lower compared to overall total of fruits exported (Jabatan Perangkaan Malaysia, 1995). Improving and optimizing the quality of carambola would increase demand by overseas markets.

Grade standards of fruit tend to be based on attributes that can be determined visually such as colour, shape, form, sizes and absence of defects. Visual grading operations use these attributes to determine acceptance or rejection of fruit for shipment to export markets (Shewfelt, 1993). The quality specifications of carambola for export are determined according to condition, cleanliness, shape and form, presence of blemishes, discoloration, damage and size of fruit (SIRIM, 1993). Skin colour, which indicates the stage of maturity of carambola, is also an important grading criterion in determining its export destination. Presently, skin colour is assessed visually by matching the fruit with a colour chart produced by the Federal Agricultural Marketing Authority (FAMA). The chart contains six colour indices representing six maturity stages (index 1= deep green, 2= light green, 3= green yellow, 4= yellow green, 5= yellow and 6= orange). Fruit of index 2 and 3 are usually exported to the European countries while fruit of index 4 and 5 are exported to Hong Kong and Singapore.

The main problem associated with the FAMA colour chart is inaccuracy in the grading system. The six colour indices of the chart do not cover the spectrum of colours that occur at the different maturity stages. Thus, determining the skin colour of a particular fruit that falls in between the indices by personal judgement becomes a difficult task during grading, resulting in a high frequency of misgrading. FAMA's six colour indices were developed by visual observation and without reference to the international colour standards such as that of the Royal Horticultural Society (RHS)

Munsell colour chart. Grading of fruit by personal judgement and without the use of any other colour measuring instruments is often flawed. Personal judgement perceived through visual images is relative from one individual to another (Judd and Wyszecky, 1975). A person's visual accuracy varies according to age, sex and light intensity (Bollen et al., 1993).

Colour-measuring instruments make it possible to determine skin colour accurately. Instruments such as the colorimeter and spectrophotometer have objective notations of colour coordinates which represent lightness, chromaticity and hue. These coordinates are used by botanists, biologists and horticulturists in the United States of America and European countries to develop colour charts based on the international colour standards, and are also widely used in evaluating or describing colour (McGuire, 1992; Voss, 1992). The coordinates obtained can be used to differentiate the skin colour changes that occur in carambola at different stages of maturity. The use of such colour measurement can avoid the effects of differences in colour perception among human observers, thus reducing human error in colour evaluation and ensuring uniformity and accuracy in colour grading.

The changes in fruit colour during ripening are the result of the loss of green colour due to the destruction of chlorophyll pigments and the presence of yellow or orange colour due to carotenoid pigments. According to Gross (1987) there are three general patterns of carotenoid development during ripening. In the first pattern the presence of yellow or orange colour is due to the degradation of chlorophyll and unmasking of the previous carotenoid pigments. Secondly, the loss of chlorophyll is followed by the synthesis of one or more carotenoid pigments and thirdly is the chlorophyll degradation followed by a decrease in carotenoid pigments. In carambola only two ripening stages have been studied, i.e., less ripe and ripe fruit whereby an increase of total carotenoids was observed (Gross et al., 1983). However, the pattern was still vague.

In addition to skin colour, changes in flesh firmness, ascorbic acid, sugar and organic acid contents, respiration rate and ethylene production at different stages of fruit maturity are important in determining quality attributes. Grade standards in the United State of America and European countries use colour and other attributes in determining their grade specifications for fresh fruits and vegetables (Kader et al., 1992). The related attributes help the consumers and producers to determine quality efficiently and quickly. However, the Malaysian Standard Specifications for Fresh Carambola (SIRIM, 1993) lacks information on such attributes. Fruits that are harvested at the early green stage usually have a firm texture but are poor in taste when ripe, whereas fruit harvested at a late stage with yellowish orange colour normally have better taste, but a shorter storage life (Rahman et al., 1992). To improve and optimise produce quality of Malaysian carambola, changes in the quality characteristics at different stages of maturity and their relationship with colour changes need to be studied. This information, once made available, could be used for better specifications for carambola and would make the Malaysian carambola more competitive in the export markets.

Therefore, the objectives of this experiment were to determine (a) the skin colour changes at different stages of maturity objectively, using a colour-measuring instrument, (b) the pigments that contribute to the skin colour and (c) changes in the quality characteristics at different stages of maturity and establish their relationships to skin colour.

CHAPTER II

LITERATURE REVIEW

Carambola

Averrhoa carambola L., which is native to Southeast Asia, belongs to the Oxalidaceae family under the order of Geraniles and the class of Dicotyledoneae in the subdivision Angiospermae of the division Spermatophyta (Hsuan, 1978). It can now be found in many parts of the globe in both large and small plantations. It is grown in Taiwan (Green, 1987), Australia (Watson et al., 1988), Florida, USA (Campbell et al., 1989; Campbell and Koch, 1987), Brazil (Donadio, 1989), Guyana (Ramsammy, 1989), Trinidad and Canary Islands (Galan-Sauco et al., 1989; Hernandez-Dolgado and Galan-Sauco, 1996), Tobago (Andrew, 1989) and Surinam (Lewis and Groeizam, 1989).

Several cultivars are grown; Arkin and Golden Star are the predominant cultivars in Florida, USA and Australia (Campbell et al., 1989; Campbell and Koch, 1987; Watson et al., 1988), Fwang Tung cultivar is commercially planted in Taiwan (Green, 1987) and Newcomb cultivar is grown in Trinidad and Canary islands (Galan-Sauco et al., 1989; Hernandez-Dolgado and Galan-Sauco, 1996). In Malaysia, seventeen cultivars of carambola have been registered (Jabatan Pertanian Malaysia, 1989). Three of these cultivars, 'B2', 'B10' and 'B17', are widely planted because of their commercial value (Rahman et al., 1992). The fruit of 'B2' is elliptical in shape and does not have a sour taste even in the immature stage. Fruit colour changes from whitish green to whitish yellow when the fruit is ripe. The fruit is about 8 - 15 cm in length, and about 200 - 250 g in weight. The flesh is juicy and has a very fine texture with soluble solids concentration (SSC) of 7 - 8%. This cultivar is normally used as a pollinator to the B10 cultivar. The B10 cultivar is about 15 - 21 cm in length and 150 - 300 g in weight. The immature fruit is green in colour and has an astringent taste. Ripe fruit is golden orange in colour and has a sweet and juicy taste (SSC of 8 - 10%). 'B17' is sweeter than 'B10' (SSC of 12 - 18%). 'B17' is distinguished from other cultivars by its elongated shape and broad winged bottom. The skin surface is freckled when the fruit is ripe (Rahman et al., 1992).

Among the tropical fruits, carambola is one of the richest in vitamins A, B1, B3 and C which are essential in the human diet (Giri et al., 1980). It is also claimed to have medicinal values for people with heart problems or high blood pressure (Padmawinata, 1980). The star-shape of carambola makes it popular in the European countries where it is mixed in fruit salad. It can be eaten fresh as a dessert or processed into juice, jam or preserves (Rahman et al., 1992).

Fruit Colour

Fruit colours are due to the chlorophylls, carotenoids and anthocyanin pigments that are present in the pulp and/or skin of fruits. The chlorophylls are green pigments that strongly absorb the red and blue regions of the visible spectrum. The small differences in the structures of chlorophyll *a* and chlorophyll *b* cause differences in the absorbtion spectra, thus producing the different green hues of the two pigments, blue-green for chlorophyll *a* (430 and 662 nm) and yellow-green for chlorophyll *b* (455 and 664 nm) (Gross, 1987). The carotenoids absorb mainly the blue region (430 - 470 nm), but they also absorb the blue-green (470 - 500 nm) and green (500 - 530) region of the spectrum and their colour is determined by the light they reflect. The hue colours they reflect increase from yellow (neurosprene) to orange and red (lycopene) (Goodwin, 1982). The anthocyanins, which appear strongly in the acid medium, have two absorption maxima, a visible region between 465 - 550 nm and an ultraviolet region of 270 - 280 nm (Gross, 1987).

For most fruits, the first visible sign of ripening is the loss of greenness. Unripe fruits contain large amounts of chlorophyll at the beginning of their development. As the fruits ripen, the chlorophylls are degraded, resulting in the loss of the green colour of the fruits. Simpson et al. (1976) reported that degradation of chlorophylls proceeds via the pheophytins or via the chlorophyllins, pheophorbides leading to tetrapyrrolic chlorins, which are eventually oxidised in light to colourless compounds. A more recently proposed model for chlorophyll degradation involves chlorophyll *a*-1 as an intermediate in addition to the cyclic tetrapyrrolic derivatives, namely chlorophyllins, pheophytins and pheophorbides. These derivatives are transformed successively into linear tetrapyrroles and dipyrroles, then into corresponding monopyrroles and linear, nitrogen-containing components and eventually into colourless simple molecules such as CO_2 and NH_3 (Hendry and Stobart, 1986). Degradation of chlorophylls during ripening has been related to the increasing activity of chlorophyllase and magnesium dechelatase enzymes (Schoch and Brown, 1987). In most fruits, the loss of chlorophylls is accompanied by the unmasking or the biosynthesis of carotenoid pigments.

Carotenoids, which are normally constituents of the photosynthetic tissue, are present in green fruit tissue prior to maturation. The first pattern of carotenoid biosynthesis takes place in the chloroplast and is an integral part of their development. It is closely linked to the biosynthesis of chloroplast components but masked by the presence of chlorophylls. During ripening the green colour turns to yellow as chlorophylls gradually degrade, unmasking the colour of previously available carotenoid pigments. The second pattern of carotenoid formations is indicated by the accumulation of large amounts of carotenoids. During ripening carotenogenic fruits gradually turn to yellow, orange or red as chlorophylls degrade and carotenogenesis takes over. This process is known as chloroplast-chromoplast transformation and is correlated with the conversion of chloroplast carotenoids in green fruits into chromoplast carotenoids by the presence of plant growth substances such as ethylene, gibberellins, abscisic acid and cytokinins. Ethylene accelerates the degradation of chlrophyll, as well as the biosynthesis of carotenoid. Gibberellins and abscisic acid reduce production of ethylene while cytokinins enhance the ability of auxin to induce ethylene production. The chromoplasts are characterised by their rapid and biosynthetic ability as reflected in the appearance of multiple carotenoids in different types (Goldschmidt, 1986). The first pattern occurs in degreening of lemon (Gross, 1987) and banana (Medlicott et al., 1992) and the second pattern occurs in tomato, Golden Delicious apples (Gross, 1992) and mangoes (Medlicott et al., 1992). In fruits such as the sweet cherry (Gross, 1982a), red currant and strawberry (Gross, 1982b) in which the final colour is imparted by anthocyanins, the total carotenoid contents decrease continuously, paralleling the decrease in chlorophylls.

In the carambola fruit (Gross et al., 1983) only two ripening stages were studied and an increase in total carotenoid level was observed. The total carotenoid content was 22 μg/g (fresh wt.). The carotenoid pattern of carambola mainly consists of phytofluene (17%), ζ-carotene (25%), β-cryptoflavin (34%) and muthatoxanthin (14%). β-carotene, β-apo-8-carotenal, criptoxanthin, cryptochrome and lutein are present in small amounts.

Visual Evaluation of Colour

Visual colour evaluation is done by matching the fruits with standardized colour charts such as the Royal Horticultural Society (RHS) or Munsell Colour charts (Voss, 1992) and more commonly by using numerical scales based on visual observation of the colour changes of the fruit. It is a widely used and accepted technique of determining quality and maturity stages of fruits, but it has many setbacks. Visual colour evaluation is subjected to variability by personal judgement. Personal judgement of visual images varies according to age, sex and light intensity. Visual acuity decreases with age, whereby a 60-year-old worker requires about twice brightness level than a 20-year-old worker (Luckiesh, 1964). Normal colour vision differs among sexes, whereby the degree of deviation is about 8% among males and 0.4% among females (Judd and Wyszecky, 1975). Changes in intensity of light alter the image received by the eye (Bollen et al., 1993).

Many colour indices have been developed to determine the maturity stages of the carambola. Oslund and Davenport (1983) divided the maturity stages of Golden Star cultivar into six colour indices according to the Munsell Colour chart (index 1 = 2.5GY 7/10; strong yellow-green, 2 = 10Y 8/11; strong green-yellow, 3 = 7.5Y 8/12; vivid green-yellow, 4 = 5Y 8/12; vivid yellow, 5 = 10YR 7/10; strong orange-yellow to 7.5YR 7/11; strong orange-yellow). Mitcham and McDonald (1991) divided 'Arkin' into four indices according to the stages of maturity (1 = Dark green,