

## EFFECT OF FLUORESCENT LIGHT ON SELECTED ANTIOXIDANT COMPOUNDS AND ANTIOXIDANT ACTIVITY DURING STORAGE OF FRESH-CUT CARAMBOLA (*AVERRHOA CARAMBOLA* L.) FRUIT

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### Abstract

The effects of fluorescent light on the availability of selected antioxidant compounds and antioxidant activity during storage of fresh-cut carambola (*Averrhoa carambola* L.) fruit were investigated. The samples of fresh-cut fruit, stored at 5 ± 1°C, were exposed to fluorescent light (157 Lux) for 12 days. Total phenolic compounds (TPC) and ascorbic acid (AA) content of the processed fruit were analyzed using Folin-Ciocalteu reagent and high performance liquid chromatography (HPLC), respectively. Antioxidant activity of the fruit was assessed following 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging capacity and ferric-reducing antioxidant power (FRAP) assays. The results of the present study showed that light exposure did not exert any significant effect, while storage period significantly reduced the AA content of the fruits tested. On the other hand, with few exceptions, there were notable random changes, recorded in the TPC and antioxidant activity of the fruit as function of storage period and light exposure.

### Introduction

The generation of free radicals in the body as a result of uncontrolled metabolism and oxidative stress is strongly linked with enhancing the incidence of degenerative disorders such as cardiovascular diseases cancer, inflammation, immune system decline and brain dysfunction (Nishikawa, 2008). Photosensitization is a widely occurring phenomenon in biological systems. Due to the ubiquitous nature of visible light together with endogenous and exogenous compounds that can act as photosensitizers, various radical and non-radical reactive oxygen substances are generated (Black, 1987). Such free radicals can accelerate skin cancer, photo aging, and other light-related pathologies (Witt *et al.*, 1993). It is widely accepted that antioxidants play an important role in protecting the body from oxidative damages and related diseases (Steinberg, 1991; Ames *et al.*, 1993; Jacob & Burri, 1996; Wang *et al.*, 1996; Maxwell & Lip, 1997; Arouma, 1998; Pratico & Delanty, 2000). Synthetic antioxidants namely butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) which are commonly used in processed foods can exert some side effects due to their perceived carcinogenic potential (Branen, 1975; Ito *et al.*, 1983). Consequently, there is a continuing interest in searching some plant-based natural and safer antioxidants due to their potential health benefits. Among the potential antioxidant sources, fruits can be an excellent source of natural antioxidants if taken regularly (Simin *et al.*, 2000; Liu, 2003; Hsin-chia *et al.*, 2004; Pierce *et al.*, 2007). Carambola (*Averrhoa carambola* L.), native to the Philippines, Indonesia, India and Sri Lanka, is popular throughout Southeast Asia, the South Pacific and parts of East Asia due to its valuable fruit. The tree is also widely grown in many parts of the tropics, such as in Colombia, Trinidad, Peru, Ecuador, Dominican Republic and Brazil, Guyana, and in the United States, in south Florida and Hawaii. Carambola can be categorized

as non-climacteric fruit (O'hare, 1993). Carambola fruit, also known as star fruit, is valued as a rich source of both primary and secondary polyphenolic antioxidants such as ((-) epicatechin and proanthocyanidins) and ascorbic acid (Shui & Leong, 2004; Shinzo *et al.*, 2008).

Many people are aware of the benefits of regular consumption of fruits. However, many do not realize that fruits, especially fresh-cut, suffer nutrients losses due to improper processing and storage conditions. Previous studies investigated the effects of different factors such as harvest season, fruit maturity, light exposure and storage conditions on the antioxidant properties of fruits (Lana & Tijssens, 2006; Shin *et al.*, 2008; Nadeem *et al.*, 2010). Shiow *et al.*, (2009) studied the effect of light and fruit maturity on the antioxidant capacity of raspberry. They reported that light enhanced raspberry's colour development, especially for the immature fruit (5 % and 20 % maturity). The researcher also found that light intensity did not affect the changes of total phenolics in 50% and 80% matured fruits during storage. This contradict with that of immature berries (5% and 20% maturity), where they reported that fruits exposed to higher light intensities had higher total phenolics in particular, during the first 2 days of storage. Light intensity was reported to exert positive effect on flavonoids content of the red raspberries. Another study by Hall *et al.*, (1994) supported the protective effect of flavonoids from light where they found that rosmariquinone, an antioxidant compound, when added in soybean oil, significantly delayed the oxidation of the oil during exposure to fluorescence light for 72 hour compared to untreated sample. Hence, it is hypothesized that fluorescence light exposure during storage can potentially affect the antioxidant compounds and antioxidant activity of fresh-cut fruit. Therefore, the present research work was undertaken with the main objective to evaluate and quantify the effects of fluorescent light on the contents of phenolics and ascorbic

acid as well as antioxidant activity of fresh-cut carambola fruits during storage.

### Material and Method

**Chemicals and reagents:** 2,2-Diphenyl-1-picrylhydrazyl solution (DPPH), 2,4,6-tripyridyls- triazine (TPTZ), acetate buffer, Ferric chloride (FeCl<sub>3</sub>.H<sub>2</sub>O), citric acid and 6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox) were purchased from Sigma-Aldrich Co. (St. Louis, USA), Methanol analytical grade was obtained from HmbG Chemical Co. (Germany), Folin-Ciocalteu reagent and Gallic acid were from Merck Co. (Darmstadt, Germany). Sodium carbonate, Na<sub>2</sub>CO<sub>3</sub> was purchased from System Co. (USA). Acetonitrile HPLC grade, methanol HPLC grade and hydrochloric acid (HCl) purchased from Fischer Scientific Co. (Leicestershire, UK).

**Sampling:** Carambola fruits cultivar B10 had been chosen because it is the most popular carambola cultivar that been planted in Malaysia. 2 kg of whole carambola fruit which consist of around 15 to 20 fruits at stage 2 maturity according to Federal Agriculture Marketing Authority standard (Anon., 2000) were purchased from wholesale market in Selangor, a day before the experiment begin and stored at temperature 5°C. Only fruit without any mechanical damages were randomly selected for the study.

**Treatment of samples:** Carambola fruit had been washed with tap water at room temperature and let to dry before been cut into dice shape with size 2 x 2 cm for equal surface area. The fresh-cut carambola had been packaged in ten food grade polypropylene (PP) plastic cases with dimension 17 x 11.5 x 5.5 cm (L x W x H) and closed with transparent plastic lids. Each plastic case contained about 90 g of sample and then divided equally into 2 different group of treatments. One group was stored in the dark while the other group been placed 200 cm under fluorescent light. Storage temperature for both conditions was 5 ± 1°C with duration 12 days. After every 3 days, one plastic case from each treated group were randomly sampled and analyzed for ascorbic acid, total phenolic compounds as well as antioxidant activity. Light intensity was measured at same distance as the sample using a photometer (model TESTO 540, Spain).

**Phytochemical extraction:** Carambola fruits were blended until homogenized before extraction by soaking in methanol with sample to solvent ratio 1: 3 (w/v) for 1 hour at temperature 40 ± 1°C (Allothman *et al.*, 2009a). The filtrate was separated from the residue by filtering through a filter paper (Whatman No. 1) and the residue reextracted again with fresh solvent according to the procedure mentioned above. The filtrates were pooled and excess methanol was then evaporated off under reduced pressure using a rotatory evaporator (Buchi Rotavapor R-210, Switzerland). The concentrated extract was then stored at - 20 ± 1°C, prior to analyses.

**Determination of ascorbic acid:** The content of ascorbic acid was determined using the method of Wimalasiri & Wills, (1983) with some modification. A weighed amount 0.5g of extract was dissolved to 20mL citric acid (3% w/v). The solution was then filtered through a filter paper (Whatman No. 1). The aqueous solution was then purified with C<sub>18</sub> Sep-Pak cartridge. The samples were filtered again using 0.45 µm filter (Whatman, Nylon Syringe Filter) and analyzed using reversed phase HPLC (LC-10 AT single pump, Shimadzu, Kyoto, Japan) utilizing µBondapak C<sub>18</sub> column (300 mm x 3.9mm i.d., 5µm). The detection was performed with UV-Visible detector (model SPD-10 AV, Shimadzu, Kyoto, Japan) at 254 nm, the mobile phase used was a mixture of acetonitrile and water (80:20 v/v) with flow rate of 1 mL/min. Retention time and peak areas of pure ascorbic acid standard were used to identify and quantify the ascorbic acid contents of the samples. The rate of AA degradation had been calculated followed the first order reaction rate. The following equation explained the degradation rate;

$$\ln C = \ln C_0 - kt,$$

where C is the concentration at time t, C<sub>0</sub> is the initial concentration and t is days of storage. The rate of degradation of ascorbic acid was determined from the gradient of the kinetic graph.

**Determination of total phenolic compounds (TPC):** The TPC were determined using Folin-Ciocalteu method as reported by Singleton *et al.*, (1999) with some modification. 0.5 mL of sample extract at concentration 1mg/mL was mixed with 0.5 mL Folin-Ciocalteu reagent, followed by addition of 10 mL of 7% sodium carbonate solution. The mixture was allowed to stand for 1 hour at 25 ± 2°C in the dark condition and then absorbance measured at 725 nm using a UV-Vis Spectrophotometer (UV-1650 PC Spectrophotometer, Shimadzu, Japan). The amount of TPC was expressed as miligram of Gallic acid equivalents (GAE) per 100g of fresh weight of sample.

### Determination of antioxidant and activity

**2, 2-Diphenyl-1-picrylhydrazyl (DPPH) scavenging assay:** DPPH assay used to determine the free radical scavenging activity of the extract was based on method of Brand-Williams *et al.*, (1995) with slight modification. The extract was diluted with methanol to prepare different concentrations in the range of 0.156mg/mL to 5mg/mL. Each dilution (0.5mL) was then added with 3.5mL of 25ppm DPPH solution. The mixture was then left in the dark at 25 ± 2°C for 30 minutes. The absorbance of the mixture was then measured at 515nm by using a UV-Vis spectrophotometer. IC<sub>50</sub> value, representing the amount of extract which scavenged/reduced 50% of the DPPH radical, was calculated from percent scavenging versus concentration curve. A higher concentration to reduce 50% DPPH solution showed lower in antioxidant activity. The calculation of % DPPH free radical scavenging was as followed:

$$\% \text{ Scavenging} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100\%$$

**Ferric reducing antioxidant power (FRAP) assay:** FRAP assay was conducted based on the method as described by Benzie & Strain, (1996) with minor modifications. The oxidant in the FRAP assay was prepared by mixing 2.5mL of 10mM TPTZ prepared in 40mM HCl, 25mL of acetate buffer, and 2.5mL of 20mM  $\text{FeCl}_3 \cdot \text{H}_2\text{O}$ . The mixture was referred to as "FRAP reagent". 200 $\mu\text{L}$  of sample was pipetted into a test tube and mixed with 3 mL of FRAP reagent by vortexing. The mixture was allowed to react for 30 minutes at temperature 37°C. Absorbance of the mixture was then read at 594nm.

**Measuring light intensity:** The degree of light emitted from fluorescence lamp to which the fresh-cut carambola fruits were exposed during this study was measured using the light meter (model TESTO 540, Spain). The intensity of the light was measured in unit of lux.

**Statistical analysis:** All analyses were performed in triplicates. Data obtained from the experiment was gathered and analyzed using SPSS 16.0 statistical programmed (SPSS Inc., Chicago, IL, USA). Analysis of Variance (ANOVA) was performed to determine significant difference of means between the control and treatments as well as among the storage periods while Pearson correlation was used to determine the significant correlation between antioxidant compounds and activity.

A probability value less than 5% ( $p < 0.05$ ) level was considered significant.

## Results and Discussion

**Total phenolic compounds and ascorbic acid in carambola:** Fresh carambola in the present study was analyzed for the contents of ascorbic acid (AA), and total phenolic compounds (TPC) and antioxidant activity utilizing 2,2-Diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging capacity and ferric ion reducing antioxidant power (FRAP) assay systems. Results of the study (Table 1) showed that ascorbic acid and TPC of carambola were found to be  $2.48 \pm 0.33$  mg per 100g FW and  $117.72 \pm 13.75$  mg GAE per 100g FW, respectively. These values were slightly lower than those reported by Lim *et al.*, (2006),  $5.20 \pm 1.90$  mg ascorbic acid per 100g FW and  $131 \pm 54$  mg GAE per 100g FW, respectively. On the other hand, Luximon-Ramma *et al.*, (2003) reported considerably higher amounts of both AA and TPC in carambola,  $14.40 \pm 3.00$  mg per 100g FW and  $209.90 \pm 10.40$  mg GAE per 100g FW, respectively. According to Shui & Leong, (2004), major phenolic compounds in carambola were identified to be (-) epicatechin and proanthocyanidins, which existed as dimers until pentamers. Shinzo *et al.*, (2008) reported the presence of epicatechin and procyanidin B2 in carambola fruits after fractionation with ethyl acetate.

**Table 1. Total phenolic compounds and ascorbic acid contents, and antioxidant activity of carambola fruit<sup>a</sup>**

Ascorbic acid content (mg/100 g FW)	Total phenolic compound (mg GAE per 100 g FW)	DPPH scavenging capacity IC <sub>50</sub> (mg/mL)	FRAP ( $\mu\text{mol TE/g FW}$ )
$2.48 \pm 0.33$	$117.72 \pm 13.75$	$1.31 \pm 0.53$	$19.78 \pm 10.44$

<sup>a</sup>Values given are means  $\pm$  standard deviation of triplicate analysis.

The variation of TPC and AA in our study compared to those reported by others may be due to different species, geographical origin and harvest time of the fruit selected (Lee & Kader, 2000; Ou *et al.*, 2002; Arshad *et al.*, 2010). Preharvest factors like climatic conditions including light and average temperature have strong influence on the chemical composition of horticultural crops (Klein & Perry, 1982). Another preharvest factor is cultural practices as studied by Lisiewska & Kmiecik, (1996) who found that amount of nitrogen fertilizer from 80 to 120 kg/ha decreased AA by 7% in cauliflower.

**Antioxidant activity of carambola:** Antioxidant activity of fresh carambola fruits as measured by DPPH assay and FRAP assay is presented in Table 1. The IC<sub>50</sub> is defined as the concentration required to reduce 50% of DPPH from the original concentration. IC<sub>50</sub> value for the fresh carambola in the present study was determined to be  $1.31 \pm 0.53$  mg/mL indicating strong antioxidant activity. Luximon-Ramma *et al.*, (2003) reported carambola antioxidant activity was  $17.00 \pm 4.00$   $\mu\text{mol}$  Trolox equivalents (TE) per g of FW as measured by ABTS assay. On the other hand, Lim *et al.*, (2006) reported that IC<sub>50</sub> of carambola fruit was  $3.80 \pm 2.10$  mg/mL using DPPH assay.

Ferric ion reducing antioxidant capacity (FRAP) is another approach widely used in determining antioxidant activity of samples. In this assay, higher absorbance means the higher amount of ferric ion being reduced by the antioxidant correlating to higher reducing power. Trolox Equivalent (TE) data was calculated based on the standard curve prepared using 6-hydroxy-2, 5, 7, 8-tetramethylchromane-2-carboxylic acid (Trolox) standard. Antioxidant activity measured by FRAP assay for the present study showed that fresh carambola exhibited  $19.78 \pm 10.44$   $\mu\text{mol TE}$  per gram of FW which is equivalent to  $495.51$  mg TE per 100g FW. Luximon-Ramma *et al.*, (2003) found that the reducing power of carambola to be  $22.00 \pm 1.00$   $\mu\text{mol Fe (II)}$  per g of FW while Lim *et al.*, (2006) using ascorbic acid to construct the standard curve, investigated FRAP value for carambola to be  $98.00 \pm 55.00$  mg AA equivalent antioxidant capacity (AEAC) per 100 g FW. It is reported that carambola contains considerable amount of primary antioxidants which can inhibit the initiation of chain reaction and thus free radical formation (Lim *et al.*, 2006). Based on the present data and the information available in the given literature, carambola can be categorized as a fruit of antioxidants with high reducing potential.

### Effect of light on selected compounds and antioxidant activity of fresh-cut carambola fruit

**Ascorbic acid:** The changes in AA content of fresh-cut carambola in relation to storage period are presented in Fig. 1. The results of the study revealed that although the ascorbic acid content of the processed fruit decreased gradually during first 12 days of storage, however no significant ( $p < 0.05$ ) difference in the content were observed between the light-exposed and control fruits. While the AA was found to be significantly ( $p < 0.05$ ) degraded as function of storage period for both the treatments. The analysis of light-exposed fresh-cut carambola revealed that the original amount of AA,  $2.49 \pm 0.50$  mg of AA per 100g FW on day 0 was decreased to  $1.15 \pm 0.43$  mg per 100g FW on day 12, a decline of 54%. On the other hand, the amount of AA of fresh-cut carambola that was stored in the dark condition, on day 12 was found to be  $1.19 \pm 0.48$  mg per 100g FW. Even though at the end of the storage period of 12 days, statistical analysis showed no significant difference for the content of AA between fresh-cut carambola stored under the light and dark condition, it is interesting to note that fresh-cut carambola stored under the dark condition undergone slower degradation compared to light-exposed fresh-cut carambola when comparing using kinetic of degradation calculation approach.

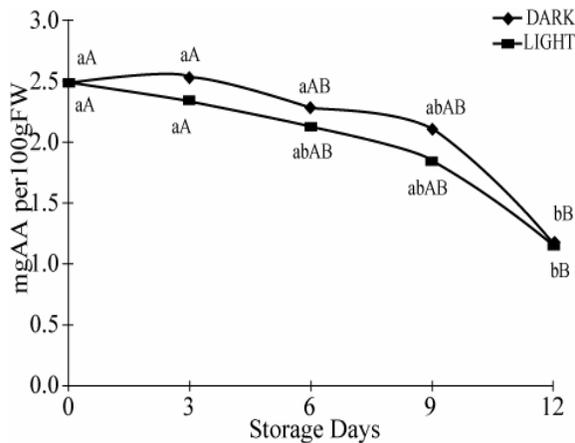


Fig. 1. Effect of light on ascorbic acid content of fresh-cut carambola during storage at 5°C

Values given are means  $\pm$  standard deviation of triplicate analysis. The values marked with same letters are not significantly different at  $p < 0.05$  analyzed using Duncan Multiple Range Test

A,B- Value with different capital letters indicated significant difference between storage days at  $p < 0.05$

a,b- Value with different small letters indicated significant difference between storage condition at  $p < 0.05$

Kinetics pattern on the rate of degradation of the ascorbic acid is shown in Fig. 2. Degradation rate of most nutrient followed zero or first order reaction, according to Labuza & Riboh, (1982). In the present experiment, fresh-cut carambola exposed to light showed higher degradation rate compared to that stored in the dark. The degradation rate,  $k$  for fresh-cut carambola stored in the dark was lower than that stored in the light with value  $0.040 \text{ day}^{-1}$

and  $0.042 \text{ day}^{-1}$ , respectively. The steeper the gradient indicated higher rate of reaction. The negative sign for gradient value only showed that the gradient was in descending pattern and thus could be ignored. Deriving the kinetics of reaction can thus predict the shelf-life of compounds in fruits during storage.

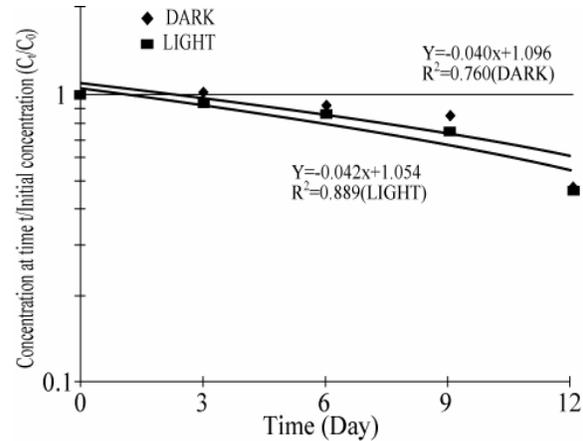


Fig. 2. Degradation kinetic of ascorbic acid in fresh-cut carambola during storage at 5°C.

Moreover, rate of a reaction depends on type of treatment applied based on previous studies. Ascorbic acid stability of orange-carrot juice treated with high pulsed electric field (PEF) during storage was investigated and compared with that of pasteurized juice (Torregrossa *et al.*, 2006). Ascorbic acid degradation rate for pasteurized juice was found to be higher than that of PEF-treated orange-carrot juice. In agreement with the present finding, ascorbic acid degradation in the given study also followed first-order reaction kinetic. Torregrossa *et al.*, (2006) found that juice stored at  $10^\circ\text{C}$  had higher ascorbic acid degradation rate compared to those stored at  $2^\circ\text{C}$ . The degradation rate of ascorbic acid for pasteurized juice stored at  $10^\circ\text{C}$  and  $2^\circ\text{C}$  were  $0.0963 \text{ day}^{-1}$  and  $0.0466 \text{ day}^{-1}$ , respectively. Ferreira & Rodriguez-Amaya, (2008) found that degradation rate for the light-exposed lycopene isolated from guava, tomato and watermelon were  $0.284$ ,  $0.387$  and  $0.270 \text{ day}^{-1}$ , significantly higher than those stored in the dark, being  $0.060$ ,  $0.148$  and  $0.086 \text{ day}^{-1}$ , respectively. Although this study using model system to simulated the nutrient in dehydrate fruit, it showed that light exposure during storage can affect the degradation rate of compounds. On the other hand, Gil *et al.*, (2006) found that the effect of light exposure to vitamin C in fresh-cut fruits varied, depending on the type of fruits. They found that fresh-cut mango and watermelon had higher vitamin C content when exposed to light while fresh-cut pineapple, strawberry and kiwi fruit showed the lower levels.

Bradley & Min, (1992) evaluated the mechanism of antioxidant destruction by light. They found that light notably affected the antioxidant compound in foods due to the presence of activating sensitizer such as chlorophyll, riboflavin and heme-containing protein. The photoactivated sensitizers can promote photooxidation, by direct interaction with an oxidizable substrate to produce free radicals. The

energy of a single photon from the light is absorbed by cell sensitizer. An electron in the absorbing molecule (chromophore) is lifted to a higher energy level (Nelson & Cox, 2005) and this will promote a reaction to occur very rapidly. It is reported that in contrast to ground-state oxygen oxidation (common triplet- oxygen oxidation, which proceeds quite slowly), the reaction of photooxidation (light-activated oxidation), proceeds about 1000-1500 times faster (Cuppert *et al.*, 1997). Light exposure can increase the rate of AA degradation since light and heat can enhance the oxidation of AA (Gregory, 1996). The results indicated that light intensity (157 lux) used in this study did not significantly affect the ascorbic acid degradation during storage. This result was in agree with Artés-Hernández *et al.*, (2010) that also did not found any significant effect of UV-C light on Vitamin C content in fresh-cut water melon. This might be due to the matrix of the fruit that acted as a shield to protect the antioxidant compound from light (Ferreira & Rodrigues-Amaya, 2008). Alothman *et al.*, (2009b) stated that in terms of exposure to radiation sources, it mainly depends on the dose applied where normally low or medium dose have insignificant effect on antioxidants. According to the study of Mitchell *et al.*, (1992), low doses irradiation on horticultural crops showed no significant effect on AA and dehydroascorbic acid (DHA) contents. Decreasing of ascorbic acid during storage may be occurred because it had been used to reduce quinones back to phenolic compounds (Teixera *et al.*, 2008).

**Total phenolic compounds (TPC):** The effect of light on TPC of fresh-cut carambola during storage is shown in Fig. 3. The result showed that up to day 5 there was no significant difference observed for TPC between the dark and light-stored carambola. On day 6, the TPC for dark and light-stored carambola were  $108.17 \pm 7.17$  mg GAE per 100g FW and  $94.37 \pm 7.89$  mg GAE per 100g FW, respectively, indicating considerable variations. At the end of storage period of 12 days, there was no statistically significant difference observed for TPC amounts in relation to condition or days of storage. The result obtained from this study showed that light and storage time did not have any significant effect on the TPC content of fresh-cut carambola.

Our findings are in agreement with Gil *et al.*, (2006) who found that fruits exposed to light did not show any significant effect on TPC when compared with those stored in the dark. As indicated earlier, the matrix of the fruit may protect the TPC from the light. The insignificant changes in TPC during storage can be supported by the study of Teixeira *et al.*, (2008) who found no significant changes on PPO activity during 9 days storage of fresh-cut carambola at temperature  $4.6^\circ\text{C}$ . As mentioned earlier, AA can play role in maintaining TPC amount by reduced quinone back into phenolic compounds after it had been oxidized by polyphenol oxidase (PPO).

The low TPC shown on day 6 may be due to oxidation of phenolic compound by polyphenol oxidase (PPO) from the loss of compartmentalization within the cells when exposed to physical or physiological stresses (Shi-Ping *et al.*, 2004). Another study by Alasalvar *et al.*, (2005) supported the increase of TPC on day 9 due to

developmental changes and wound like responses. Accumulation of phenolic compounds occurred because of physiological response to infectious or injuries (Amanatidou *et al.*, 2000). Wounding stimulates phenylalanine ammonia lyase (PAL, E.C. 4.3.1.5) activity that increases the phenolic compounds (Salveit, 1997). PAL was activated by phenylpropanoid metabolism because of the induced reactive oxygen species (Reyes *et al.*, 2006). This enzyme can function to deaminate the respective amino acids into phenolic acid such as cinnamic and *p*-coumaric acids (Boudet, 2007). Both enzymes are absent in healthy tissues and their rapid formation in cut and infected tissues is dependent on protein synthesis. Guillermo *et al.*, (2001) found PAL activity in carambola increased until day 10 when stored at temperature 2 and  $10^\circ\text{C}$  because of the chilling injury response which increased the TPC. Besides that, the increment of the TPC after day 9 also can be contributed by the water loss during storage of fresh-cut carambola at  $5^\circ\text{C}$  (Ali *et al.*, 2004).

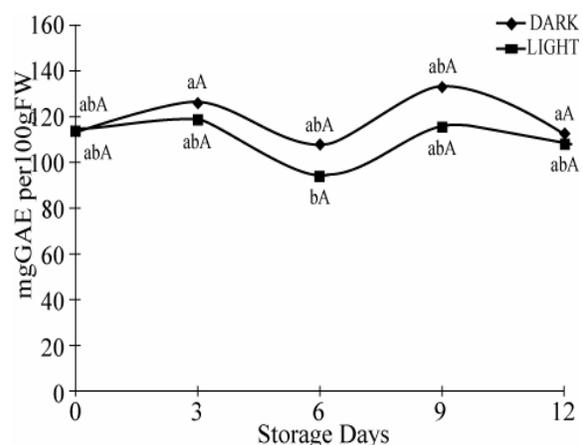


Fig. 3. Effect of light on total phenolic compounds in fresh-cut carambola during storage at  $5^\circ\text{C}$ .

Values given are means  $\pm$  standard deviation of triplicate analysis. The values marked with same letters are not significantly different at  $p < 0.05$  analyzed using Duncan Multiple Range Test

A, B- Value with different capital letters indicated significant difference between storage days at  $p < 0.05$

a, b- Value with different small letters indicated significant difference between storage condition at  $p < 0.05$

**Antioxidant activities:** Effects of light on the antioxidant activity of carambola as measured by DPPH and FRAP analysis are shown in Figs. 4 & 5, respectively. There was no significant ( $p < 0.05$ ) difference observed for the DPPH radical scavenging capacity between dark and light-exposed samples (Fig. 4). However, the scavenging ability was noted to be decreased on day 6 for both samples with  $\text{IC}_{50}$  value of  $1.08 \pm 0.11$  mg/mL and  $1.14 \pm 0.15$  mg/mL, respectively. Eventually, the radical scavenging capacity of the fruits tested increased on day 9 with  $\text{IC}_{50}$   $0.84 \pm 0.10$  mg/mL and  $0.86 \pm 0.10$  mg/mL, respectively. This  $\text{IC}_{50}$  value increased until day 12 with contribution for dark and light-exposed sample of  $0.65 \pm 0.01$  and  $0.73 \pm 0.09$  mg/mL, respectively. The trend for radical scavenging capacity was similar to those seen for

TPC as discussed earlier with significant ( $p < 0.01$ ) correlation coefficient value 0.551. It is worth pointing out that FRAP assay may also measure other phenolic compounds that not readily react with DPPH solution like phenol, *p*-coumaric acid, vanillin, vanillic acid and  $\gamma$ -resorcylic acids (Brand-William *et al.*, 1995; Bondet *et al.*, 1997). The results of the study showed no significant ( $p < 0.05$ ) difference in the antioxidant activity between carambola stored in the light or dark conditions (Fig. 5). The lowest FRAP value,  $23.88 \pm 3.69 \mu\text{mol TE per g FW}$  and  $21.57 \pm 4.11 \mu\text{mol TE per g FW}$  were observed on day

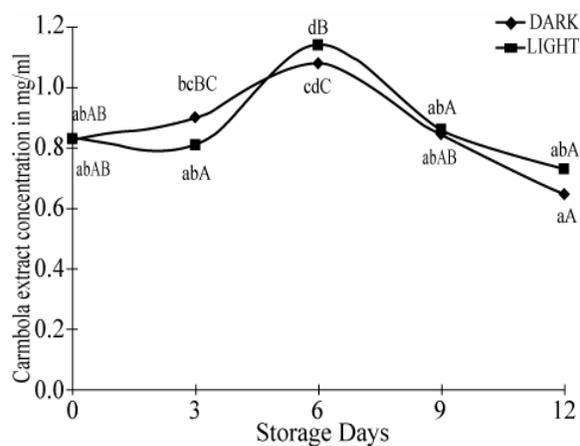


Fig. 4. Effect of light on DPPH scavenging activity ( $IC_{50}$ ) of fresh-cut carambola during storage at  $5^{\circ}\text{C}$ .

Values given are means  $\pm$  standard deviation of triplicate analysis. The values marked with same letters are not significantly different at  $p < 0.05$  analyzed using Duncan Multiple Range Test

A,B- Value with different capital letters indicated significant difference between storage days at  $p < 0.05$

a,b- Value with different small letters indicated significant difference between storage condition at  $p < 0.05$

The result of the present study agreed with that of Wicklund *et al.*, (2005), where no significant ( $p < 0.05$ ) difference in the FRAP value between strawberry jam, stored under fluorescence light and dark condition, during storage for 3 month were reported. They also found that antioxidant capacity was affected more by storage temperature rather than exposure to fluorescence light. However, they found low correlation between FRAP value and colour values. This occurred because the antioxidant capacity may be contributed by colourless phenolic compounds based on Jiratanan & Liu, (2004) study. In contrast, according to Artés-Hernández *et al.*, (2010) fresh-cut watermelon exposed to UV-C light for 11 days had high antioxidant activity. They suggested this increment occurred due to stress induced by UV-C light

6 for the fruits stored under dark and light conditions, respectively. According to FRAP assay the antioxidant activity of carambola fruit, exposed to both the storage conditions, was significantly ( $p < 0.01$ ) correlated with TPC of the sample ( $R^2 = 0.685$ ). This can be explained by the low TPC value on day 6 discussed previously. Similarly, on day 12, antioxidant activity of the fruit exposed to light was lower than that of the sample stored in the dark, FRAP value of  $25.96 \pm 3.23 \mu\text{mol TE per g FW}$  and  $29.29 \pm 6.25 \mu\text{mol TE per g FW}$ , respectively.

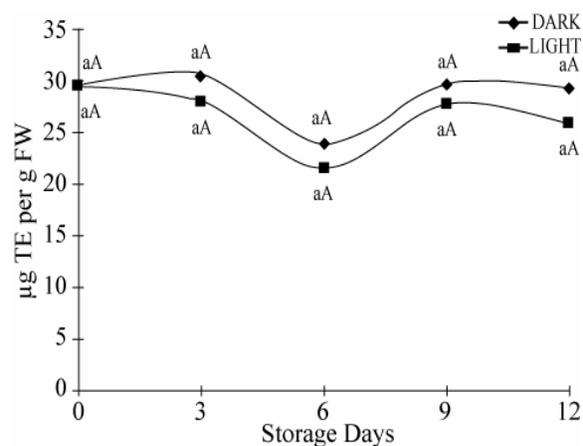


Fig. 5. Effect of light on ferric reducing antioxidant power (FRAP) of fresh-cut carambola during storage at  $5^{\circ}\text{C}$ .

Values given are means  $\pm$  standard deviation of triplicate analysis. The values marked with same letters are not significantly different at  $p < 0.05$  analyzed using Duncan Multiple Range Test.

A,B- Value with different letters indicated significant difference between storage days at  $p < 0.05$

a,b- Value with different letters indicated significant difference between storage condition at  $p < 0.05$

treatment and storage. Kalt *et al.*, (1998) reported that phenolic composition, anthocyanins, and antioxidant capacities in strawberries, raspberries and high bush and low bush blueberries changed during postharvest storage, and consequently affected the antioxidant capacity of these fruits. Overall, in the present study, antioxidant activity of carambola was strongly influenced by TPC as compared to that of ascorbic acid content as showed in Table 2. Shui & Leong, (2002) reported that ascorbic acid had poor contribution towards antioxidant activity of certain fruits. Besides, Piga *et al.*, (2002) also found that changes in the antioxidant properties of fresh-cut mandarin were weakly linked to ascorbic acid concentration.

**Table 2. Correlation of total phenolic compounds and ascorbic acid with antioxidant activity of carambola fruit during storage as measured by DPPH and FRAP assays.**

	DPPH	FRAP
Ascorbic Acid	0.059	0.347
Total Phenolic Compound	0.551**	0.684**

\*\* The values marked are significantly different at  $p < 0.01$  using Pearson correlation.

## Conclusions

Selling of fresh-cut fruit is a new trend in fruit marketing and based on fruit nutritional shelf-life, fresh-cut carambola is suitable for fresh-cut fruit market. The results of the present investigation revealed that light exposure at intensity 157 lux exhibited minimal effect on the contents of total phenolic compounds, ascorbic acid, and antioxidant activity of fresh-cut carambola fruits. The effects of light during storage on the total phenolic compounds and antioxidant activity of the fruit were insignificant ( $p < 0.05$ ). However, considerable degradation in ascorbic acid was still observed at day 12 for the fruits exposed to both the light and dark conditions, revealing significant ( $p < 0.05$ ) difference between the storage days. Carambola fruit's antioxidant properties were mainly contributed by phenolic compounds as evident by correlation data.

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