

CHANGES IN PHYSICO-CHEMICAL TRAITS AND ENZYMES OXIDATIVE SYSTEM DURING COLD STORAGE OF 'FORMOSA' PAPAYA FRESH CUT FRUITS GROWN IN THE MEDITERRANEAN AREA (SICILY)

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ABSTRACT

In this study, the effects of cold storage ($5\pm 0.5^{\circ}\text{C}$ and relative humidity of $90\pm 1\%$) on the quality of fresh papaya slices packed in a passive atmosphere with a semi-permeable film were evaluated. Physico-chemical traits such as total soluble solids, reducing sugar, pH increased during storage as well as the polyphenols, carotenoid content and antioxidant activity that reaching the highest values at end of trials. Changes in colorimetric parameters resulted in a significant decrease after 4 days of hue angle values, which then remained constant. The cutting process enhanced the antioxidant enzymes activity such as superoxide dismutase, catalase and ascorbate peroxidase. The analysis of the main components showed physical-chemical, qualitative, and enzymatic changes in papaya samples during cold storage, showing a shift from negative to positive values along the PC1 and indicating a qualitative decay of sliced papaya.

Keywords: papaya, minimally processing, enzymes, color, antioxidant, packaging

1. INTRODUCTION

Papaya (*Carica papaya* L.) is the fifth most widely produced tropical fruit worldwide after mango, banana, pineapple, and avocado, with 13.0 million tons per year (LIU *et al.*, 2019). Papaya is a perennial herbaceous plant recently spread in Spain and Italy (FARINA *et al.*, 2020a), where, in recent years, papaya has adapted to the Mediterranean climate under sheltered structures (FARINA *et al.*, 2020b) with good quality results (FARINA *et al.*, 2020a), like other new crops (NIRO *et al.*, 2017). Its cultivation is supported by a constant demand for freshly cut papaya, especially in Europe, mainly to young consumers and "baby boomers", who eat it as a snack (JAMES *et al.*, 2010). Papaya is a type of climacteric fruit whose maturation after harvest is accompanied by tissue softening and microbial growth (GONZALEZ-AGUILAR *et al.*, 2009). The proximity to European markets makes it possible to harvest the climacteric fruits close to their ripening stage, allowing for excellent organoleptic attributes (GENTILE *et al.*, 2019) that reduce the number of kilometers of food and greenhouse gas emissions. In this context, the search for methods that use simple operations and equipment to improve the shelf life of minimally processed papaya is of interest to farmers and consumers (JAYATHUNGE *et al.*, 2014). Several studies have demonstrated an increase in the reactive oxygen species (ROS) level after the cutting process in fresh fruit such as carrot (JACOBO-VELÁZQUEZ *et al.*, 2011), *Zizania latifolia* (LIU *et al.*, 2012), and pitaya (LI *et al.*, 2017). ROS have harmful effects in various cellular compartments at high levels, but these compounds may also act as signaling molecules in ripening and senescence processes in response to cutting stress (JACOBO-VELÁZQUEZ *et al.*, 2011). However, the balance between ROS generation and scavenging is closely modulate by antioxidant defense system characterized by enzymatic and non-enzymatic components in fresh-cut fruit (HODGES *et al.*, 2008). Different enzymes control the intracellular metabolism of ROS, such as superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione peroxidase (GPX) and catalase (CAT), which modulate the ROS level. The cutting process induces an increase of ROS above threshold levels, with damages to cell membranes due to lipid peroxidation with a high lipoxygenase activity and malondialdehyde content (KARAKURT and HUBER, 2003; SOUZA *et al.*, 2015; WU *et al.*, 2019). Furthermore, in fresh cut fruit have been observed a high enzymatic browning due to loss of cellular compartmentalization that allows to the polyphenol oxidases (PPOs) come in contact with phenolic compounds causing fruit color changes (WU *et al.*, 2019). The aim of this research was to study the effect of cold storage (5°C and RH of 90±1%) and passive atmosphere packaging on the shelf-life of minimally processed papaya fruit grown in Mediterranean climate under greenhouse, by monitoring the variations in physico-chemical, microbiological and nutraceutical traits. In addition, in order to assess the physiological stress induced by the cutting and slicing process, the enzymatic oxidative system and the markers of oxidative damage have also been studied.

2. MATERIALS AND METHODS

2.1. Fruit samples and experimental design

'Formosa' papaya fruits were picked in a commercial orchard in Palermo, Italy (33S 0333746 m.E, 4217131.00 m.N). The fruits were harvested at stage 3 (one or more orange-colored stripes in skin; pulp almost completely orange in color, except near skin, still hard but contains less latex), using the skin color of the fruit as a maturity index (BASULTO *et*

et al., 2009). The subsample of 25 fruits (5 per tree) was selected. Fruits were stored before the evaluation under a controlled temperature (18°C, 90% RH) and analyzed when papayas reached the stage 4 (BASULTO *et al.*, 2009) after 3 days. 9 subsamples of the fruit were submitted to physical-chemical determination whereas 6 subsamples of the fruit were washed with cold tap water and peeled with the help of a stainless-steel knife; the seeds were removed, and the fruits were cut into small pieces approximately 2.5 cm thick. About 120 g of the cut samples were placed inside sanitized plastic trays (142 x 95 x 50 mm) and wrapped with a film Cryovac Sealappeal PSF. The characteristics of the film are as follows: thickness 25 µm; transmission rate of CO₂ 800 cm³m⁻²day⁻¹ at 23°C and 0% RH; transmission rate of O₂ 72 cm³m⁻²day⁻¹ at 4°C and 0% RH; moisture transmission vapor 47 gm⁻²day⁻¹ at 38°C and 100% RH. Three replicates were prepared for each sampling time (9 packages) that were stored for 12 days at 5°C and RH of 90±1%. Samples were analyzed every four days and for each biological replicate were realized two technical replicates. The chosen time of storage adopted in this study emerged from preliminary assays performed to determine the probable shelf life resulted in early mold formation on the 12th day at 5°C (data is not shown).

2.2. Determination of physico-chemical traits

2.2.1 Skin color (raw fruit)

For the skin cover color index (CC) evaluation (at harvest and consumption points), we used the fruit analysis system (FAS) procedure in agreement with FARINA *et al.* (2011).

2.2.2 Flesh color

Chromaticity values L* (Lightness), a* (green to red), and b* (blue to yellow) of flesh color fruit was determined with a colorimeter (Minolta C2500, Konica, Ramsey, NY), Chroma (Chr) and hue angle (Hue) were also calculated (FARINA *et al.*, (2020c).

2.3. Physico-chemicals analyses

A digital scale (Gibertini, Italia) was used for determining the fresh weight (FW) and the size code was determined (CBI, 2018). Flesh firmness (FF) was measured using a digital penetrometer TR5325 (Turoni, Forlì, Italy) with a cylindrical needle (8 mm diameter) and values expressed in Newtons (N). Flesh juice was used to detect the total soluble solids content (TSS) with an optical digital refractometer (Atago, Japan), titratable acidity (TA) using a compact titrator (Crisom, Spain) and expressed in g citric acid/100 g fresh fruit and pH with a digital pH-meter (Model 2001, Crison, Barcelona, Spain). Reducing sugars were evaluated through a volumetric Fehling assay as described previously by ADILETTA *et al.* (2018).

2.4. Carotenoids, polyphenols content and antioxidant activity

Spectrophotometric detection of carotenoids content, xanthophylls plus carotenes (CAR), was determined in agreement with PETRICCIONE *et al.* (2015) and results were expressed as µg 100 g⁻¹ FW applying Wellburn equations (WELLBURN, 1994). The total phenolic compounds (TP) were assessed as reported by MAGRI *et al.* (2020). The free radical

scavenging activity was gauged by 1,1-diphenyl-2-picryl-hydrazil (DPPH) according to CINQUANTA *et al.* (2013).

2.5. Evaluation of antioxidant enzymatic system

Total soluble proteins were obtained from fresh papaya (1:3 w/v) blended in an extraction buffer prepared in agreement with MAGRI *et al.* (2020) and determined by the Bradford assay. Catalase activity (EC 1.11.1.6) (CAT) was estimated in according to the method described by ADILETTA *et al.* (2019) using 100 μL of crude enzyme extract. The activity was expressed in μmol of H_2O_2 g^{-1} FW. Superoxide dismutase activity (EC 1.15.1.1) (SOD) was evaluated with the method of nitroblue tetrazolium (NBT) reduction inhibition in agreement with ADILETTA *et al.* (2018a) using 50 μL of crude enzyme extract. SOD activity was expressed as U mg^{-1} FW, considering that one SOD unit corresponds to the amount of enzyme that, in the assay conditions, inhibits 50% the NBT reduction. Guaiacol peroxidase activity (EC 1.11.1.7) (GPX) was determined according to PETRICCIONE *et al.* (2015) and expressed as nmol g^{-1} FW. Ascorbate peroxidase activity (EC 1.11.1.7) was assessed as reported by ADILETTA *et al.* (2018b). The activity was expressed as μmol g^{-1} FW. Hydrogen peroxide content was estimated with the method reported by GOFFI *et al.* (2020) and expressed as nmol g^{-1} FW.

2.6. Oxidative damage markers and enzymatic browning

Polyphenoloxidase activity (EC.1.10.3.1) (PPO) was established with the method described by ADILETTA *et al.* (2018b) and PPO assay was carried out with 10 μL of crude enzyme extract. PPO activity was expressed as μmol g^{-1} FW. Malondialdehyde content (MDA) was determined as described by ADILETTA *et al.* (2018b) and expressed as nmol g^{-1} FW. Lipoxigenase activity (EC 1.13.11.12) (LOX) was determined in according to ADILETTA *et al.* (2018b) and expressed in nmol $\text{m}^{-3}\text{g}^{-1}$ FW, as the specific rate of molar change of hydroperoxides.

2.7. Microbiological analysis

Total aerobic bacteria (TAB) and yeast and mold count (YM) were conducted using a method designated by YOUSUF and SRIVASTAVA (2015). Results were reported as log colony forming units per gram.

2.8. Statistical analysis

Analysis of variance (ANOVA) and Duncan's test at a 5% level were used to compare the differences between samples analysed during storage. Principal components analysis (PCA) was realized to reduce the multidimensionality of dataset generating new principal components that account for most of the total variation. All statistical analyses were realized by SPSS software package, Version 20.0 (SPSS Inc., Chicago, IL, USA).

3. RESULTS AND DISCUSSION

3.1. Fresh fruits

The examined fruits were classified in H size (1101 - 1500 g) in according to CBI Size Codes A–J used for marketing channels (CODEX STAN, 2001). Consumers tend to prefer smaller papayas, particularly in northwestern Europe; because they suit individual consumption better and consider the fresh-cut papaya more convenient than the whole fruit (which should be peeled, deseeded and sliced before consumption) and they find the large size of some cultivars off-putting (RIVERA-LÓPEZ *et al.*, 2005). Fruit with <55% yellow skin was the best to slicing and deseeding while those with <25% yellow skin showed no soft and edible flesh. In this study, papaya fruit were harvested at mature green stage 3 (BASULTO *et al.*, 2009) and was characterized by some orange-colored stripes in skin; pulp almost completely orange in color, except near the skin, but still hard for consumption. In this stage, the color of the skin changes from dark green to light green and one or more yellow streak begins to develop from the base upwards. The cover color analysis revealed 33.3% of skin cover color at picking; afterwards fruits were let to ripe at room temperature reaching 59.6 % of yellow skin color after 4 days. Our results agree with YAHIA (2011), who suggested to select whole fruit with 55-80% of yellow skin which ensures > 50% of edible flesh recovery for production of fresh-cut papaya because fully ripe fruit were easily bruised and difficult to handle YAHIA (2011). Nonetheless, after these storage conditions the fruit reached good physico-chemical characteristics for fresh-cut processing (Table. 1). L^* , chroma and hue values were comparable with those reported by GAYOSSO-GARCÍA *et al.* (2011) in the flesh of raw papaya fruit and by FARINA *et al.* (2020b) in fresh-cut coated fruit; similar TSS, TA (ZUHAIR *et al.*, 2013) and pH values were observed by ZUHAIR *et al.* (2016). Finally, flesh firmness values were in agreement with other studies carried out on papaya fruit at the same ripening stage grown in tropical (FARINA *et al.*, 2020a) and Mediterranean area (FARINA *et al.*, 2020b).

3.2. Physico-chemical traits in fresh-cut fruits

Formosa papaya has fruit with a large size and orange flesh, for these features are generally valued as minimally processed fruits. In papaya fruit, flesh colour changes can be attributed to different regulation of carotenoid biosynthesis, a secondary metabolic pathway that yield metabolites also destined to other important fruit qualitative features (YAHIA, 2011; SHEN *et al.*, 2019). Furthermore, the increase in metabolic activities also causes flesh colour changes during fresh-cut processing (RIVERA-LÓPEZ *et al.*, 2005). The intensity and uniformity colour of flesh fruit affect its quality and consumer's choice and preference (RIVERA-LÓPEZ *et al.*, 2005). Samples showed a significant decrease in L^* after 4 days of cold storage, followed by a constant trend up to 12 days (Fig. 1A). Furthermore, the results showed a significant increase in redness a^* (from 46.3 to 52.3) and decrease in yellowness b^* (from 56.2 to 48.6) after 12 days of storage (Figs. 1B and C). WAGHMARE and ANNAPURE (2013) observed a similar trend in passive atmosphere packaging, and JAYATHUNGE *et al.* (2014), using micro-perforated polyvinyl chloride containers. As a result, changes in colorimetric parameters resulted in a significant decrease after 4 days of hue angle values, which then remained constant (Fig. 1D), while no significant differences were registered in chroma values during storage conditions (data not shown). In other word, red colour increased its intensity in all samples during the storage period; this was due to the degradation of chlorophylls or unmasking of preformed pigments during fruit

development. The colour changes ranged from yellow to reddish orange and were associated with ripening progress, as well as at the onset of browning. TSS in fresh-cut papaya tended to increase during cold storage with significant differences during storage time (Table 1). The highest TSS (8.51%) value in papaya samples was found on day 12. WAGHMARE and ANNAPURE (2013) reported a similar trend in TSS, explaining it by the solubilisation and synthesis of carbohydrates in fresh-cut papaya packed in polypropylene film with and without modified atmosphere and stored for 25 days at 5°C. Moreover, the low temperature helped to maintain a low level of respiration rates stopping the decrease in TSS content (RIVERA-LÓPEZ *et al.*, 2005). Reducing sugar slightly increased in fresh-cut papaya during storage showing a high positive correlation ($R^2=0.969$ $p < 0.01$) with TSS (Table 2). The pH values increased significantly during storage, ranging from 5.51 to 5.64 after 12 days of cold storage.

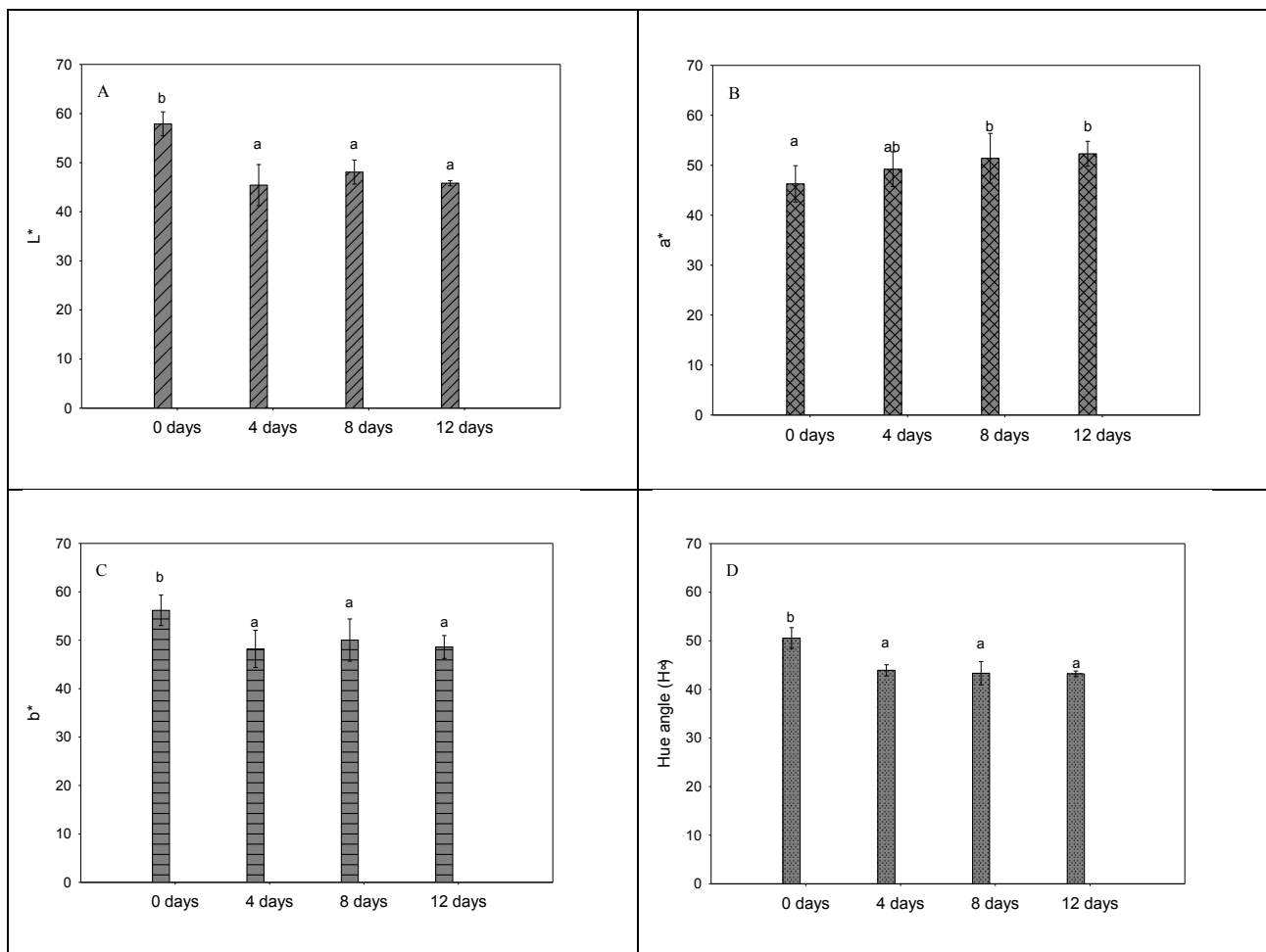


Figure 1. Evaluation of colorimetric traits, lightness L^* (A), redness index a^* (B), yellowness index b^* (C) and Hue angle H° (D) in cut 'Formosa' papaya fruit packaged in passive atmosphere stored at 5°C for 12 days. Means \pm standard deviations followed by the same letter do not differ significantly at $P = 0.05$ (Duncan Test).

Table 1. Pomological traits of the *Formosa* papaya cultivar after three days of ripening at room temperature (18°C - RH 90%). Values represented as mean \pm SD (n=9). Fruit weight (FW), Solid soluble content (TSS), Titratable acidity (TA), Firmness (F), Juiciness (J), skin cover color (CC), Lightness(L*), Chroma (Chr), hue angle (Hue).

FW (g)	TSS (Brix°)	TA (g/L)	F (N)	J (ml/100 g)	CC (%)	L*	Chr	Hue
1380 \pm 20	8.03 \pm 0.9	0.8 \pm 0.21	33.42 \pm 3.8	46.21 \pm 2.6	59.6%	62.85 \pm 7.9	60.96 \pm 7	1.02 \pm 0.3

Table 2. Changes in total soluble solid (TSS), reducing sugar (RS) and pH in cut 'Formosa' papaya fruit packaged in passive atmosphere stored at 5°C for 12 days.

Storage Time	TSS	RS	pH
Time 0	8.03 \pm 0.02 a	2.23 \pm 0.11 a	5.51 \pm 0.01 a
4 days	8.02 \pm 0.02 a	2.29 \pm 0.22 b	5.56 \pm 0.01 ab
8 days	8.09 \pm 0.01 b	2.42 \pm 0.13 c	5.59 \pm 0.01 bc
12 days	8.51 \pm 0.01 c	2.79 \pm 0.41 d	5.64 \pm 0.01 c

Means followed by the same letter do not differ significantly at P = 0.05 (Duncan Test).

3.2.1 Microbial growth in cut fruit

Fresh-cut fruit is more prone to the rapid growth of spoilage microorganisms as well as the pathogens of public health significance. Peeling process eases cross-contamination and the transfer of microflora from peel to the flesh fruit that represents an optimal substrate for microbial growth. Cold storage of fresh-cut papaya leads to an increase in TAB and YM values. TAB increased in all stored samples ranging from 1.8 (0 day) to 5.6 log CFU/g (12 days) (data not shown). These values were lower than the critical limit for total microbial loads of vegetables (8.0 log CFU/g) (JACXSENS *et al.*, 2002). YM values increased from 1.3 to 5.2 log CFU/g, overcoming the critical limits of 5 log CFU/g for yeasts (JACXSENS *et al.*, 2003) after 12 days of storage. Increased trend in microbial counts of TAB and YM throughout the storage of fresh-cut papaya were also reported by Gonzalez-Aguilar *et al.* (2009) and WAGHMARE and ANNAPURE (2013), correlated to the packaging systems, storage temperatures and different cut types of fresh-cut fruits.

3.2.2 Free radical scavenging power components in fresh cut fruits

Papaya is a tropical fruit with a high concentration of bioactive compounds such as polyphenols, vitamins and carotenoids, whose interactions contribute to the overall antioxidant activity of this fruit. Carotenoids are a group of fat-soluble molecules responsible to yellow-red color of fruits and vegetables. Ripening, cold storage and postharvest treatments can influence carotenoids content in fresh-cut fruit (SUPAPVANICH *et al.*, 2020). The samples tested showed an increase in the carotenoids content during storage time, ranging from 570 \pm 12 (0 days) to 1600 \pm 92 μ g 100 g FW⁻¹ (12 days) (Fig. 2A). Our results were in agreement with FAJAR FALAH *et al.* (2015) who evaluated fresh-cut 'Bangkok' papaya at different storage temperatures suggesting that this climacteric fruit continues to ripe during storage. In addition, it can also be assumed that carotenoids are more extractable due to changes in cell structure during storage time. TP

content significantly increased during 12 days of cold storage in fresh-cut 'Formosa' papaya ranged from 19.1 ± 1.8 (0 day) to 54.3 ± 4 (12 days) mg GAE 100 g^{-1} FW (Fig. 2B). Wounding stress caused by cutting process might contribute to an increase of secondary metabolites such as phenolic compounds. Several studies have evaluated TP content in different papaya fruit highlighting that several factors such as fruit ripening, agronomic practices and post-harvest storage conditions affect the content of these bioactive compounds (ALI *et al.*, 2014; ZUHAIR *et al.*, 2013). Throughout the 12 days of cold storage, the DPPH radical-scavenging activity significantly increased ($p < 0.05$) in stored samples (Fig. 2C). Bioactive compounds can act as antioxidants and our results indicated that high antioxidant activity in fresh-cut papaya fruit was related to the increase of polyphenols ($R^2 = 0.840$; $p < 0.01$) content due to ripening process occurring during storage.

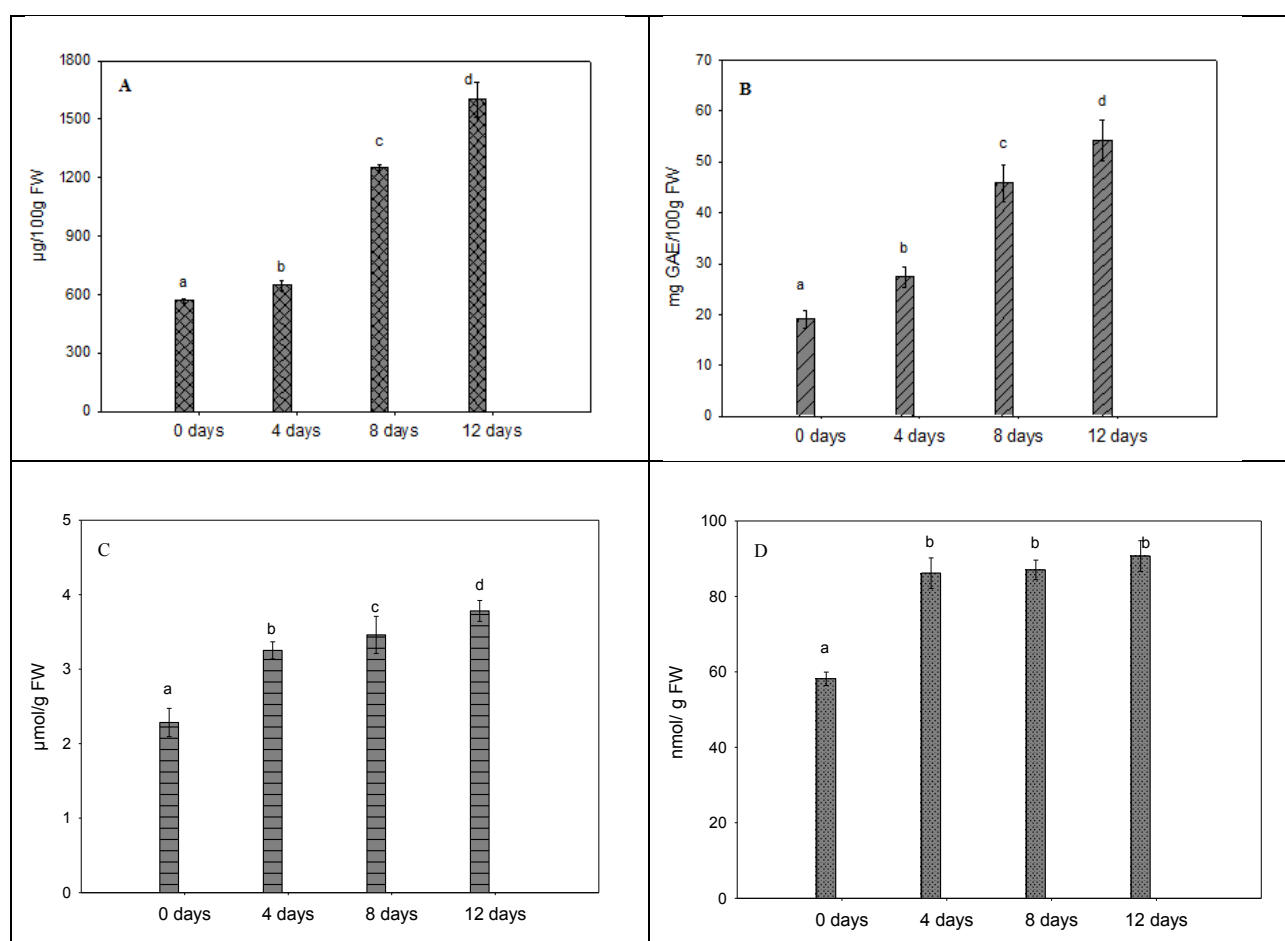


Figure 2. Carotenoids content (A; $\mu\text{g } 100 \text{ g}^{-1}$ FW), polyphenols content (B; mg GAE 100 g^{-1} FW), antioxidant activity (DPPH assay) (C; $\mu\text{mol TE g}^{-1}\text{FW}$), malondialdehyde content (D; nmol g^{-1}FW) in cut 'Formosa' papaya fruit packaged in passive atmosphere stored at 5°C for 12 days. Means followed by the same letter do not differ significantly at $P = 0.05$ (Duncan Test).

3.2.3 Antioxidant system in cut fruits

The stress by cutting process induces a physiological disorder with an alteration in cellular homeostasis increasing the ROS production in damaged cells. In fresh-cut fruit, the imbalance between production and accumulation of ROS could be due to enhancing the respiration rate and activation of amine- and NADPH-oxidases (MITTLER, 2002). H_2O_2 level increased rapidly during the first 4 days from cutting process and this trend continued up to 12 days of storage. H_2O_2 acts as a second messenger for the induction of several defense genes in crops in response to wounding (Table 3). Our results highlighted marked changes in enzymatic oxidative system due to the exposure to wounding stress in fresh-cut fruit. The superoxide dismutase (SOD) activity of fresh-cut papaya showed a slow increase through storage time, with minimal significant changes from 15.4 ± 2 U mg^{-1} FW (0 day) to 20.2 ± 4 U mg^{-1} FW (12 days) (Table 3). During the first 8 days of storage, the ascorbate peroxidase (APX) activity had no significant changes until 8 days, with an average value of 0.87 ± 0.12 $\mu mol g^{-1}$ FW, while its activity significant increased at the end of storage (12 days) (Table 2). A significant increase was registered in catalase (CAT) activity that during storage time up to 14.9 ± 2 $\mu mol g^{-1}$ FW (Table 3). Our results suggest that an increase of activities of antioxidant enzymes such as SOD, CAT, and APX, could improve the ability of the fresh-cut fruit to dismutate superoxide radicals and to eliminate hydrogen peroxide. In papaya, these enzymes prevent oxidative damage and reduce the susceptibility to chilling injury at low-temperature storage (HANIF *et al.*, 2020). In cut fruit, antioxidant enzymes can modulate ROS levels and CAT and APX activities increased when ROS reached toxic levels. At low and moderate levels, ROS can act as signaling molecules and in cut fruit such as pitaya and strawberry mediating wounding-induced phenolic accumulation (LI *et al.*, 2017; JACOBO-VELAZQUEZ *et al.*, 2015).

3.2.4 Oxidative damage in cut fruits

The cut fruit showed a high perishability due to the peeling and cutting processes that caused cell disruption and membrane damage with decompartmentation of cellular structures, cellular functions and quality loss (PAL *et al.*, 2004; JACOBO-VELAZQUEZ *et al.*, 2015). In cut tissues, several enzymes come into physical contact with their substrates afterwards the cellular damages due to the cutting process (KARAKURT and HUBER, 2003). Browning is the result of enzymatic oxidation of phenolic compounds in lightly processed fruit (JACOBO-VELAZQUEZ *et al.*, 2015). Polyphenoloxidase (PPO) and GPX are the main intracellular oxidative enzymes involved in enzymatic oxidation in stored fresh-cut fruit. PPO and GPX activities increased significantly during storage in the fresh-cut papaya up to 2.5- and 3.1-fold, respectively at the end of the experiment (Table 3). This suggests that the browning of fresh-cut papaya is primarily due to phenolic compounds oxidation caused by PPO and GPX activities. In the cut papaya, fruit lipoxygenase (LOX) activity significantly increased throughout cold storage with values ranging from 12.4 ± 1 nmol g^{-1} FW (0 days) to 36.2 ± 3 nmol g^{-1} FW (12 days) (Table 3). Instead, MDA content increased rapidly (47 %) during the first 4 days and then was stable around 88.9 ± 5 nmol g^{-1} FW up to the end of storage (Fig. 2D). As suggested by KARAKURT and HUBER (2003), LOX activity is involved in peroxidative lipid metabolism with a possible relationship with tissue softening in fresh-cut and whole 'Sunrise Solo' papaya during cold storage. Several studies have demonstrated that LOXs are ripening-related enzymes in papaya fruit (FARINA *et al.*, 2011). Our results confirm that in fresh-cut papaya the membrane lipid peroxidation occurred during cold storage. MDA content is useful to

evaluate the cell oxidative damage during storage and the effectiveness of post-harvest treatments in several fresh-cut fruit samples (GONZALEZ-AGUILAR *et al.*, 2009; SOUZA *et al.*, 2015; JACOBO-VELÁZQUEZ *et al.*, 2011).

Table 3. Evaluation of superoxide dismutase (SOD; U mg⁻¹ FW), catalase (CAT; μmol g⁻¹ FW), ascorbate peroxidase (APX; μmol g⁻¹ FW), guaiacol peroxidase (GPX; nmol g⁻¹ FW), polyphenoloxidase (PPO; μmol g⁻¹ FW), lipoxygenase (LOX; nmol g⁻¹ FW) activity and H₂O₂ content (nmol g⁻¹ FW) in cut 'Formosa' papaya fruit packaged in passive atmosphere stored at 5°C for 12 days.

Storage Time	SOD	CAT	APX	GPX	PPO	LOX	H ₂ O ₂
Time 0	15.4±2a	6.1±0.9a	0.8±0.1a	39.9±3a	0.4±0.05a	13±1.2a	0.02±0.01a
4 days	16.8±3ab	8.9±1b	0.9±0.1a	82.7±2b	0.4±0.1b	20±2.1b	0.06±0.01b
8 days	19.6±3ab	12.9±1c	0.9±0.1a	97.6±2c	0.6±0.1c	31±2.9c	0.11±0.03c
12 days	20.2±4c	14.9±1d	1.3±0.2b	124.8±7d	1.1±0.12d	39±3.8d	0.11±0.02c

Means followed by the same letter do not differ significantly at P = 0.05 (Duncan Test).

3.3. PCA in cut fruit

A dimensional map with loadings and scores plot obtained by PCA analysis is shown in Fig. 3.

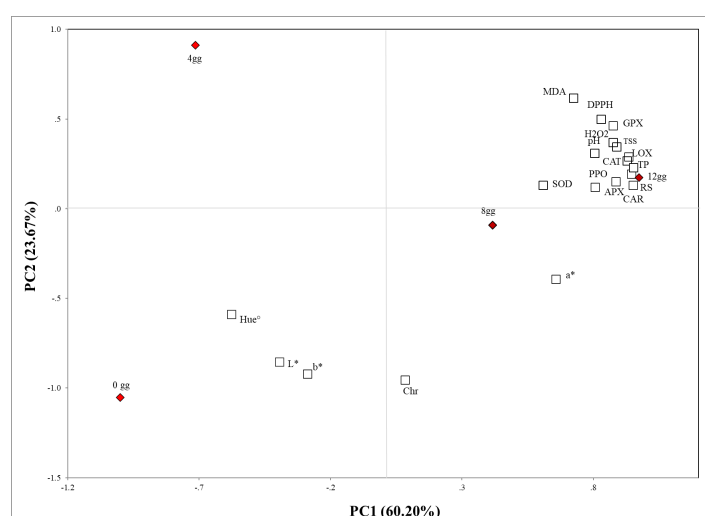


Figure 3 Principal component analysis of the physico-chemical, nutraceutical, and enzymatic traits in cut 'Formosa' papaya fruit packaged in passive atmosphere stored at 5°C for 12 days. (TSS: total soluble solid; pH; L*: lightness; a*: redness; b*: yellowness; HUE: Hue angle; Chr: chroma; TP: total polyphenol content; CAR: carotenoid content; DPPH: antioxidant activity; SOD: superoxide dismutase, CAT: catalase, APX: ascorbate peroxidase; PPO: polyphenoloxidase; GPX: guaiacol peroxidase; LOX: lipoxygenase; MDA: malondialdehyde content; H₂O₂: hydrogen peroxide content.

The first two principal components (PCs) explained 60.20% and 23.67% of the total variance, respectively. TSS, pH, a*, CAT, GPX, SOD, APX, PPO, LOX, H₂O₂ content, TP and CAR were positively correlated to PC1. PC2 showed a positive correlation with MDA and DPPH and a negative correlation with L*, b* and HUE. A shift along the first two PCs of score values highlighted physico-chemical, qualitative and enzymatic changes in fresh-cut papaya samples during cold storage.

At the beginning of cold storage, fresh-cut samples were situated in III quadrant after four days a shift from negative to positive ones along PC2 was registered. As cold storage progressed, samples displayed a shift from negative values to positive ones along PC1 showing qualitative decay of the fresh-cut papaya. The quality of fresh-cut papaya fruit, which includes physico-chemical, microbiological and nutritional traits, was preserved for 8 days of cold storage, while oxidative damages and qualitative decay were observed as the storage period progressed (12 days).

4. CONCLUSIONS

The semi-permeable packaging and cold storage (5±0.5°C) have extended the post-harvest period up to 12 days of minimally processed 'Formosa' papaya, preserving its microbiological and qualitative decay. Physico-chemical and nutritional traits changed as storage progressed due to the physiological processes associated with the ripening stage in papaya fruit. Cold storage leads to an increase in total aerobic bacteria, with lower values to the critical limit for total microbial loads of vegetables (8.0 log CFU/g) while the yeast and mold count slightly exceeded (5.2) the critical limit of 5 log CFU/g after 12 days of storage. Cutting process improved the enzymatic antioxidant system to modulate the ROS level. Enzymatic browning markers highlighted that a progressing oxidative damage occurred during cold storage. The balance between scavenging and production of ROS by the enzymatic antioxidant system allows regulating of ROS content that could be important signaling molecules in mediating the wound-induced bioactive compounds accumulated in fresh-cut papaya fruit during storage.

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