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### Commercial potential of fruits from albus varietal groups of chayote

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#### **Abstract**

Mexico has the greatest variability of chayote (*Sechium edule* Jacq. Sw.) fruits. Nevertheless, there are no reports on postharvest treatments for *albus*-type white chayotes. The objective was to evaluate different postharvest treatments in the *albus* varietal groups: *a. minor*, *a. levis*, *a. dulcis*, *a. spinosum*, and *a. levis gigante*, harvested in Huatusco, Veracruz, Mexico. To do this, the fruits were stored at 12 ±1 °C and 90 ±5% RH for seven days and then kept at 18 ±1 °C and 60 ±5% RH for evaluation. The treatments were wax, plastic film, citric acid and ascorbic acid, alone or in combination. Weight loss, commercial quality, enzyme activity, membrane permeability, and phenol content were determined. It was observed that the *albus*-type fruits are sensitive to epicarp darkening, weight loss, viviparity, and incidence of fungi, with a shelf life of between 1 and 6 days. The use of wax coating and plastic films, individual or combined with organic acids, reduced weight losses and the enzymatic activity of polyphenol oxidase (PPO) and peroxidase (POD), increasing the shelf life of the fruits by 6 to 12 days.

### **Keywords:**

chayote varietal groups, darkening, Quality.



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## Introduction

Mexico is the center of origin of chayote [Sechium edule (Jacq.) Sw.], so it has the greatest diversity of this species in the world. Currently, 12 varietal groups are recorded, which are differentiated based on their morphological, biochemical, and genetic characteristics. Within these varietal groups, yellow chayotes of the albus type are the following: a. minor, a. levis, a. levis gigante, a. dulcis, and a. spinosum, which distinguish themselves by the yellow epicarp and the slightly sweet taste of their pulp (Iñiguez-Luna et al., 2021).

Compared to other varietal groups, *albus* chayotes contain a higher amount of total soluble solids (TSS, 7.21-8.08%) and lower titratable acidity (0.28-0.035%) (Cadena-Iñiguez *et al.*, 2011). Likewise, *albus* chayotes are an important source of bioactive metabolites: for example, *a. minor* and *a. levis* have 16.16 and 10.48 mg g-1 dry weight (d.w.) of cucurbitacins plus 1.64 and 1.59 mg g-1 d.w. of flavonoids, respectively (Iñiguez-Luna *et al.*, 2021).

In Mexico, the distribution of *albus* groups is restricted to areas with an altitude between 1 160 and 2 398 m and only in the states of Veracruz, Puebla, Hidalgo, and Oaxaca. In this sense, it is estimated that by 2050, *a. dulcis* will lose more than 50% of its current distribution due to prolonged droughts and rising temperatures. In addition, in their natural habitats, *albus* groups are gradually being replaced by the introduction of more commercial chayote varieties, such as smooth-skinned green chayote (*virens levis*) (González-Santos *et al.*, 2017).

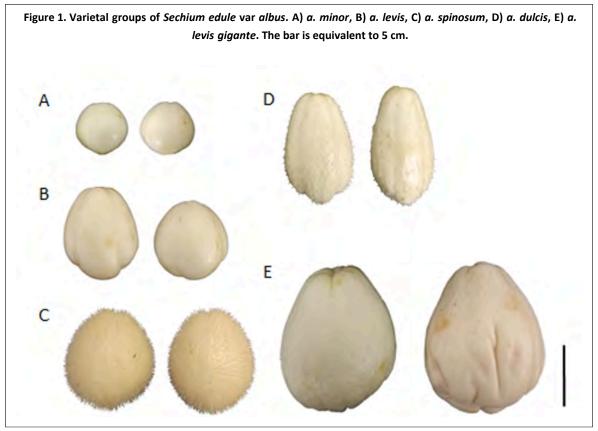
This has caused the *albus* groups to be highly susceptible to extinction, which would lead to losing part of the genetic diversity of the species *S. edule*. To avoid this, the establishment of commercial crops of *albus* groups has been initiated. Nonetheless, there are no reports that show the postharvest management strategies so that the fruits can be marketed beyond local markets.

Refrigeration is a widely used postharvest technique; however, low temperatures can lead to cold damage (CD). For example, in cucurbits, such as cucumber (*Cucumis sativus*), storage at 4 °C caused the darkening of the epidermis due to increased electrolyte leakage and changes in membrane fatty acids, damaging the appearance of the fruit (Zhang *et al.*, 2023). In this regard, applications of organic acids combined with modified atmospheres (MA) have had positive effects in delaying the darkening of fruits; for example, the use of xanthan gum-based coatings (10% w/v) combined with ascorbic acid (1% w/v) or citric acid (1% w/v) more than halved the PPO activity of grape fruits, var. *Pinot noir*, after 21 days of storage at 4 °C (Golly *et al.*, 2019).

Similarly, the application of ascorbic acid (40 mmol L-1) and oxalic acid (2 mmol L-1) combined with controlled atmospheres (5% CO2+1% O2) reduced epicarp darkening in lychee fruits by reducing membrane ion leakage and PPO and POD activities and increasing the activity of antioxidant enzymes, such as catalase (CAT) (Ali *et al.*, 2021). This study aimed to evaluate the effect of citric acid and ascorbic acid, alone and incorporated together with plastic films, on the darkening of the epicarp and the commercial quality of *albus* chayote fruits stored in refrigeration.

### Materials and methods

Chayote fruits from the varietal groups: a. levis, a. dulcis, a. minor, a. levis gigante, and a. spinosum, were obtained in November 2021 from the National Germplasm Bank of Sechium edule in Mexico (BANGESe, for its acronym in Spanish) located in Huatusco, Veracruz, Mexico (19° 08' 48" north latitude and 97° 57' 00" west longitude). Fruits without physical damage or pests were selected and harvested at horticultural maturity (18 ±2 d after anthesis). The fruits of a. minor had weights from 12 to 27 g; a. levis from 51 to 148 g; a. dulcis from 43 to 108 g; a. spinosum from 162 to 287 g, and those of a. gigante, from 168 to 374 g (Figure 1).



The fruits of each varietal group, washed and selected, were divided into four treatments. The acid solution included the combination of citric acid (0.2% w/v) + ascorbic acid (1% w/v) in distilled water. The immersion time of the fruits was 5 min. The treatments were as follows: T0= control; T1= PHS Clarity® wax, sprayed directly on the fruits; T2= immersion in acid solution; T3= plastic film (perforated polyethylene used by producers, 27 µm thick) on *a. spinosum* and *a. levis gigante*. In the fruits of *a. minor*, *a. levis*, and *a. dulcis*, sealed polypropylene bags (thickness 31 µm) were used; T4= immersion in acid solution and subsequent application of PHS Clarity® wax and T5= immersion in acid solution and plastic film.

After the application of treatments, all fruits were stored in refrigeration (12 ±1 °C and 90 ±5% RH) for seven days, followed by storage at temperatures of 18 ±1 °C and 60 ±5% RH simulating marketing conditions. The application of treatments depended on the number of fruits available; thus, for *a. spinosum*, all treatments were applied, whereas for the rest of the varietal groups, only T0, T4, and T5 were applied. One fruit was considered per replication and each treatment had 15 replications.

Weight losses were determined with a digital balance (Setra® model SI-2000S, USA) with a sensitivity of 0.01 g. The results were expressed as a percentage of weight loss with respect to the initial weight of the fresh fruit. Epicarp darkening was classified based on the following levels: level 0= no spotting, 1= mild; 2= moderate; 3= severe. The levels of viviparity were as follows: level 0= no seed, 1= visible seed and basal opening and 2= fully exposed seed. The shelf life ended when the fruits showed: weight loss greater than 10%, dehydration, viviparity, oxidation of the epidermis, and fungal damage.

Due to the availability of *a. spinosum* fruits, three replications were performed per treatment for the enzymatic activity, total phenols, and membrane permeability. Enzyme activity was evaluated in acetone powder from the epidermis of the fruits (Alia-Tejacal *et al.*, 2005). Eight grams of pericarp was taken and homogenized with 16 ml of cold acetone (-4 °C) in a blender for 1 min. The macerate was then filtered, the process was repeated two more times, and the acetone powder was left to dry at room temperature (21 °C) and stored in freezing at -4 °C until use.



PPO activity (EC 1.10.3.1) was evaluated with the method reported by Lamikanra (1995). At a room temperature of 19 °C, 3 ml of catechol (60 mM) dissolved in a Tris-HCl buffer (100 mM, pH 7.1) and 0.2 ml of the enzymatic extract obtained from acetone powder were mixed. The mixture was stirred and the absorbance change was measured at 420 nm on a UV-Vis spectrophotometer (Thermo ScientificTM, model Genesystm 10UV). Enzymatic activity was reported as U g-1 of fresh weight, where one unit of activity is equal to the formation of 1 μmol of o-benzoquinone min-1.

POD activity (EC 1.11.1.7) was measured by following the method described by Flurkey and Jen (1978). For this purpose, 2.6 ml of the Tris-HCl buffer (100 mM, pH 7.1), 0.25 ml of 0.1 M guaiacol, 0.1 ml of 0.25% hydrogen peroxide, and 0.05 ml of the enzymatic extract obtained from acetone powder were taken. The absorbance change was determined at 470 nm in 3 min in a UV spectrophotometer (Thermo ScientificTM, model Genesystm 10UV). One unit of activity is equal to the formation of 1 µmol of tetraguaiacol min-1 (U g-1 fresh weight).

CAT activity (EC 1.11.1.6) was evaluated as described by Martínez-Damián *et al.* (2013). Three milliliters of Tris-HCI buffer (10 mM, pH 8.5) and 0.1 ml of 0.88% hydrogen peroxide were taken in 100 mM of Tris-HCI and mixed in a quartz cell. The reaction was initiated by adding 0.1 ml of the enzymatic extract, observing the change in absorbance at 240 nm in a UV spectrophotometer (Thermo ScientificTM, model Genesystm 10UV). For enzymatic activity, one unit of activity is equal to the breakdown of 1 µmol of H2O2 min-1 (U q-1 fresh weight).

Total phenols (TP) were obtained by following the methodology by Singleton *et al.* (1999). Samples were read at 765 nm in a UV spectrophotometer (Thermo ScientificTM, model Genesystm 10UV). The results were expressed as equivalents of µg gallic acid per g of fresh weight.

Membrane permeability was evaluated as described by Qu *et al.* (2009). Three chayote pericarp discs were extracted, 1 cm in diameter and 4 mm thick. The discs were washed three times with deionized water and placed in 30 ml of mannitol (0.2 M). After stirring for 2 h at room temperature, the initial electrical conductivity was measured with a digital conductivity meter (Beckman 4.0, New York, USA). The discs were then boiled for 5 min and the total electrical conductivity was measured. Electrolyte leakage was expressed as a percentage of total conductivity.

The analysis was performed with the R software (version 4.0.2). A completely random design was used. An analysis of variance and a multiple comparison of means (Tukey, #= 0.05) were performed.

### **Results and discussion**

Treatments with citric acid and ascorbic acid plus wax (T4) and both acids plus film (T5) significantly reduced weight loss in all varietal groups (Table 1). T5 fruits reduced weight loss by 91, 92, 95, and 48% for *a. minor*, *a. levis*, *a. dulcis*, and *a. levis gigante*, respectively. The fruits of the *albus* varietal group lose a lot of weight, particularly those of *a. minor*.

Table 1. Quality of *albus* fruits: *a. minor*, *a. levis*, *a. dulcis*, and *a. levis gigante*, after refrigerated storage (1 week at  $12 \pm 1$  °C and  $90 \pm 5\%$  RH) and 4 days at  $18 \pm 1$  °C and  $60 \pm 5\%$  RH.

Varietal group	Treatment	Weight loss (%)		Darkening		Shelf life (d)
		Upon exiting refrigeration	Day 4	Upon exiting refrigeration	Day 4	_
T4	4.43 b	13.32 b	0.36	1.45	3	
T5	0.52 a	0.84 a	0	0.18	6	
a. levis	TO	4.31 b	17.86 c	1.5	2	3
	T4	3.85 b	9.16 b	1.16	1.66	5
	T5	0.34 a	0.69 a	0.17	0.33	10
a. dulcis	ТО	4.8 b	16.54 b	1.36	2	3
	T4	4.36 b	14.14 b	1.2	1.7	6



Varietal group	Treatment	Weight loss (%)		Darkening		Shelf life (d)
		Upon exiting refrigeration	Day 4	Upon exiting refrigeration	Day 4	_
a. levis gigante	TO	1.93 b	6.09 b	0.83	1.83	6
	T4	1.92 b	4.61 ab	0.66	1	10
	T5	1 a	3.66 b	0.5	0.83	12

Where: T0= control; T4= citric acid (0.2%) and ascorbic acid (1%) + wax and T5= citric acid (0.2%) and ascorbic acid (1%) + plastic film. Different letters within each day indicate significant differences between treatments (Tukey, p < 0.05, n= 10).

This weight loss was attributed to the metabolic processes of perspiration and respiration; in chayote *var. virens levis*, there are reports of perspiration values between 1.1 and 2.7 g kg-1 h-1 at harvest time (Ramírez-Rodas *et al.*, 2023) and respiration between 0.86 and 1.3 µg kg-1 s-1 (Cadena-Iñiguez *et al.*, 2006). However, the *albus* fruits experience between 54 and 191% greater daily weight loss than *virens levis*, except for *a. levis gigante*, which loses less weight (Rivera-Ponce *et al.*, 2024).

The use of modified atmospheres was effective in reducing the darkening of the fruits, with plastic films being more efficient, in combination with organic acids (Table 1). Ascorbic acid is able to prevent melanin formation by binding to intermediate metabolites, whereas copper chelating agents, such as citric acid and oxalic acid, are able to suppress PPO activity by binding to metal cofactors in the PPO enzyme (Golly *et al.*, 2019).

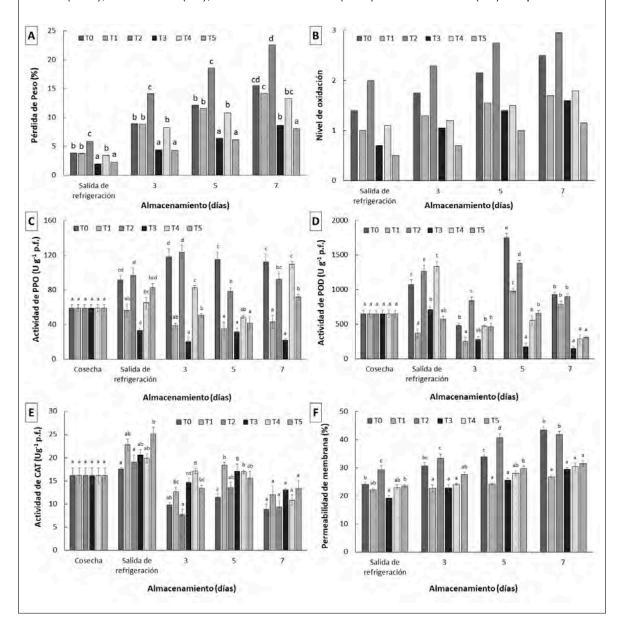
Shelf life increased significantly in all T5 varietal groups. It is important to note that the high relative humidity around the fruits with plastic film caused the development of fungi from day 6 in *a. minor* and day 10 in *a. dulcis* and *a. levis* (Table 1). The shelf life of the control fruits of *a. spinosum* was four days; for its part, the use of plastic films was the most effective in reducing weight loss and extending shelf life for an additional six days (Figure 2A). The positive effect of using plastic films has been observed in other cucurbits. In cucumber, the low-density polyethylene (LDPE, 37.5 µm) reduced weight loss by more than 65% at 12 days of storage at 10 °C and 85% RH (Kaur *et al.*, 2021).





Figure 2. Effect of treatments on weight loss (%), oxidation level (0 - 3), enzymatic activity (U g-1 f.w.), and membrane permeability (%) in fruits of *S. edule* var. *albus spinosum* stored for 1 week at 12  $\pm$ 1 °C and 90  $\pm$ 5% RH, followed by storage at 18  $\pm$ 1 °C and 60  $\pm$ 5% RH (mean  $\pm$  standard error).

T0= control; T1= wax (PHS Clarity®); T2= citric acid (0.2%) and ascorbic acid (1%); T3= plastic film; T4= citric acid (0.2%), ascorbic acid (1%), wax and T5= citric acid (0.2%) and ascorbic acid (1%) and plastic film.





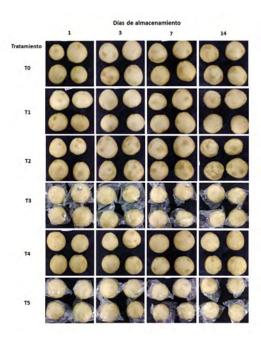
In zucchini (*Cucurbita pepo*), polyvinyl chloride film reduced fruit weight loss by about 70% at 15 d of storage at 10 °C and 80% RH (Guerra *et al.*, 2020). Once the fruits were moved to room temperature, the darkening became more apparent, but was mitigated by plastic film and wax (Figure 2B). PPO activity in the epicarp increased after transferring the fruits to room temperature, with maximum activity on the third day of storage. The fruits from T0 and T2 showed the greatest epicarp darkening and the highest PPO and POD activities until the fifth day of storage (Figure 2C and 2D).

CAT activity was higher in treatments that had less oxidation of the epidermis. That is, wax, plastic film, and combination with acids (p < 0.0001). Nevertheless, the activity is steadily reduced from the moment the product leaves refrigeration and during storage in all treatments (Figure 2E). As for viviparity (early germination of the seed), it began at two days of storage in environmental conditions in *a. spinosum*. In the control fruits, it occurred in 50%, while in the fruits of the treatments, it occurred in 30%.

At harvest, the TP content in the pericarp of *a. spinosum* was  $407.64 \pm 16.75 \,\mu g$  g-1. Nonetheless, at the end of refrigeration, it increased in all treatments, being lower in T3, with  $484.11 \pm 11.93 \,\mu g$  g-1, and higher in T2, with  $639.02 \pm 22.61 \,\mu g$  g-1, which is related to the darkening of the fruits (Figure 3).

Figure 3. Appearance of fruits of chayote *albus spinosum* stored for one week at 12  $\pm$ 1 °C and 90  $\pm$ 5% RH, followed by storage at 18  $\pm$ 1 °C and 60  $\pm$ 5% RH for 14 days.

T0= control; T1= wax (PHS Clarity®); T2= citric acid (0.2%, w/v) and ascorbic acid (1%, w/v); T3 = plastic film; T4= citric acid (0.2%, w/v) and ascorbic acid (1%, w/v) plus wax; T5= citric acid (0.2%, w/v) and ascorbic acid (1%, w/v) plus plastic film.



This increase can be attributed to cold stress, which induces the expression of genes related to the biosynthesis of phenylpropanoids, precursors of phenolic compounds (Liu *et al.*, 2024). In addition, when loss of membrane integrity occurs, the enzymes come into contact with the substrate, causing darkening (Figure 3). Changes in permeability lead to an increase in POD activity and ROS formation, resulting in peroxidation of membrane phospholipids (Luo *et al.*, 2021). In pear (*Pyrus bretschneideri* Rehd), the use of LDPE-based films (10 µm) reduced the expression of genes related to PPO biosynthesis (Cheng *et al.*, 2015).

### **Conclusions**

The fruits of *albus* chayotes are susceptible to rapid weight loss, epicarp oxidation, viviparity, and fungal incidence in refrigerated and room temperature storage. The problems of weight loss and darkening of the fruit pericarp can be mitigated with the use of waxes and plastic films, individual or combined with organic acids, because they reduce the oxidative activity of PPO and POD and increase the activity of CAT, with the consequent increase in the shelf life of the fruits. Therefore, using plastic films and waxes are a viable option for preserving the quality of *albus* fruits in refrigerated and room temperature storage.

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