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Full Length Research Paper

# Effects of boiling and oil or vinegar on pickled jurubeba (Solanum paniculatum L.) fruit

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Jurubeba fruit, raw and thermally processed for different periods of time, were preserved in soybean oil or vinegar and evaluated for physical characteristics, phytochemicals, antioxidant capacity and polyamines. The loss of green color in many vegetables after cooking is a frequent problem that affects the quality of pickled foods, and chlorophyll content is a relevant parameter to assess the quality. Data showed that a 20 min cooking treatment maintained the best fruit quality and no change in the chlorophyll content occurred. The thermal processing caused no increase in the carotenoid and flavonoid content as compared to the raw fruits, but caused an increase in the phenol content. At a cooking time of about 10 min, the antioxidant capacity increased. Cooking time did not cause significant differences in the content of isoorientin, rutin and caffeic acid. Spermine and spermidine contents were lower after 20 min of boiling. Jurubeba that was preserved in vinegar showed a lower pH and putrescine level, regardless of the cooking time used, whereas the use of oil caused an increase in carotenoids and antioxidant capacity.

**Key words:** Thermal processing, antioxidants, polyamines, phytochemicals, Solanaceae.

#### INTRODUCTION

Globally, significant improvements have been made through studies of regional dietary habits and considerable inter- and intra-country variability (Kearney, 2010). Much important information has been provided by research that has focused on non-conventional foods that are consumed together with staple foods to improve taste and nutritional quality. These non-conventional foods include *Solanum paniculatum* L. (Solanaceae), popularly

known as "Jurubeba", which is widely used in folk medicine as a tonic and antipyretic agent (Santos et al., 1988). This plant is native to northern and northeastern regions of Brazil and produces a dark-green fruit that is used for culinary purposes.

S. paniculatum fruits are mainly consumed after cooking, with rice or as pickles prepared either in oil or vinegar. Heat treatment is the main cause of change in

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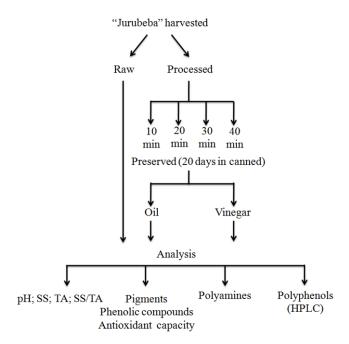


Figure 1. Representative flow chart of fruits preparation and analysis performed

the content of natural antioxidants in food (Kaur and Kapoor, 2008). However, the cooking process can contribute to the formation of new compounds, or promote the easier extraction of molecules from the cell matrix. In fruits and vegetables, many bioactive compounds, such as polyphenols, carotenoids and polyamines, are present in variable concentrations. The plants of the genus Solanum contain steroidal saponins, glycoalkaloids and flavonoids- secondary metabolites that are important for the natural defenses of plants (Ramos et al., 2012). An evaluation of some plants in relation to their antioxidant potential (the chemical constituents that assist in freeradical scavenging), such as polyphenols, vitamins, alkaloids, triterpenes, sesquiterpenes and other molecules has been the object of several studies (Ramos et al., 2012).

The most common polyamines (PAs) in fruits and vegetables are putrescine, spermidine and spermine-compounds that are frequently affected by the cooking process and heat treatments (Rossetto et al., 2015). Some fruits are rich in putrescine (Lima et al., 2008), whereas green vegetables are richer in spermidine (Valero et al., 2002). The occurrence of polyamines in jurubeba has not yet been described and their quantification is necessary.

Information regarding the nutritional composition of *S. paniculatum* fruits, *in natura* or after cooking, is scarce or absent. Thus, the aim of this research was to evaluate the effects of thermal processing on the quality of *S. paniculatum* fruits by assessing the content of vitamin C, pigments, total phenols, total flavonoids and characterizing

some polyphenols by HPLC, and analyzing total antioxidant capacity in raw and heat-treated samples. In addition, the effect of preservatives such as oil or vinegar on pickled fruits was established.

#### **MATERIALS AND METHODS**

# Reagents and samples

All chemicals and flavonoids standards (isoorientin, rutin or caffeic acid) were purchased from Sigma Co. (St Louis, MO, USA).S. paniculatum fruits were harvested in February 2014, from a farm located in Caceres, Mato Grosso State, Brazil, (16°04'14"S latitude, 57°40'44"W longitude and 118 m altitude), and were transported to the laboratory. This species was identified by the botanical characteristics and a voucher (BOTU 027535, BOTU 027536 and BOTU 027537) was previously deposited at the UNESP Herbarium, in Botucatu, São Paulo State, Brazil (Vieira Junior et al., 2015). A homogeneous and representative sample, whole fruits with intact peel and in the same maturity stage, was selected and sanitized in a commercial chlorine bleach solution.

#### Cooking process and the preparation of pickles

To obtain cooked samples, 150 g fruit was placed in stainless steel pans with 1 L boiling distilled water and was cooked for 10, 20, 30 or 40 min at atmospheric pressure. After this procedure, the remaining water was drained and the fruits were cooled at room temperature.

Raw and cooked fruits were pickled in sterile flasks filled with 2.5% NaCl in two different preservatives: soybean oil or alcohol vinegar. After preparation, the pickle bottles were sealed and stored at room temperature  $(23 \pm 2^{\circ}C)$  for 20 days, as shown in Figure 1.

#### Soluble solids, pH and titratable acidity

The soluble solids were analyzed using a digital refractometer (Atago, PAL-1 model). The pH was determined by a potentiometer (model Q Quimis - 400A). Titratable acidity was determined in gram citric acid 100 g<sup>-1</sup>, by titration, using 2 g of ground product and 20 mL of distilled water, as previously reported (Bett-Garber et al., 2015).

#### Vitamin C

The amount of vitamin C was determined using 2 g fruit ground in liquid nitrogen and diluted in 10 mL oxalic acid. This method is based on the reduction of 2,6-dichlorophenolindophenol dye by ascorbic acid (Zenebon et al., 2008).

#### Carotenoids, chlorophyll, total phenols and flavonoids

Raw, cooked and pickled samples were ground with liquid nitrogen and stored at -80°C, until analysis. The determination of carotenoids and chlorophyll (a and b) was carried out using the method of Sims and Gamon (2002). From each sample, 100 mg was homogenized in a mini-turrax (Marconi, Brazil) with 3 mL cold acetone/Tris-HCl (0.2M, pH 7.8, 80:20, v/v) solution for 1 min. All procedures were conducted on ice and were protected from light. The samples were then centrifuged at 2,000 g for 5 min and the supernatant was immediately used for the determination of pigments, using a UV/VIS spectrophotometer (Amersham-Pharmacia-Biotech). Total chlorophyll was obtained by the sum of chlorophyll a and b. The absorbance values were converted into  $\mu g$  total carotenoids. $g^{-1}$  based on the formula:

Carotenoids =  $\{A_{470}-[17.1^*(Cla+Clb)]-9,479^*anthocyanin\}/119.26^*$ Chlorophyll  $a = 0.01373^*(A_{663})-0.000897^*(A_{537})-0.003046^*(A_{647})$ Chlorophyll  $b = 0.02405^*(A_{647})-0.004305^*(A_{537})-0.005507^*(A_{663})$ .

Total phenols were determined based on the method of Singleton and Rossi (1965) using the Folin-Ciocalteu reagent. Fresh powdered samples (100 mg) were homogenized in a mini-turrax (Marconi, Brazil) with 5 mL acetone: water (50:50 v/v). After 20 min in an ultrasonic bath (Eco-sonics, Ultronique), samples were centrifuged for 10 min at 6,000 g at 5°C. The supernatant was removed and retained. The extraction process was performed once and the supernatants were combined. Absorbance was measured at 725 nm and the results were expressed in mg gallic acid equivalents  $\rm g^{-1}$  fresh weight.

For flavonoid analysis, the samples were extracted in methanol. After 60 min in an ultrasonic bath, the samples were centrifuged at 6000 xg (HeitichZentrifugen, MIKRO 220R) for 10 min and the supernatant was collected. Extraction was conducted in the pellet twice and supernatants were combined and analyzed for the total flavonoid (Popova et al., 2004). The complete process was carried out in the absence of light. The results were expressed in mg quercetin g<sup>-1</sup> fresh weight.

#### Trolox equivalent antioxidant capacity (TEAC) assay

The antioxidant capacity was determined using the methodology proposed by Brand-Williams et al. (1995), adapted by Rossetto et al. (2009). The results were expressed in uM of Trolox equivalents µg g<sup>-1</sup> sample (TEAC). The extract was obtained from 100 mg pulverized fresh samples in liquid nitrogen, homogenized in a miniturrax (Marconi, Brazil) and kept for 15 min in an ultrasonic bath (Eco-sonics, Ultronique) with 3 mL ethanol. Subsequently, the samples were centrifuged for 10 min at 6,000 g at 5°C and after 30

min, the absorbance was measured at 517 nm, in a UV/VIS spectrophotometer (Amersham-Pharmacia-Biotech).

### Thin layer chromatography of polyamines (PAs)

Polyamines were analyzed as described by Flores and Galston (1982), with modifications by Lima et al. (2008). Solanum paniculatum fruits were homogenized in cold perchloric acid (5% v/v) for 1 h and were centrifuged (10000 xg, HeitichZentrifugen, MIKRO 220R) for 30 min at 4°C. Then, 4.5 mol L<sup>-1</sup> Na<sub>2</sub>CO<sub>3</sub>, containing 18.5 mmol L<sup>-1</sup> dansyl chloride in acetone (Sigma, 95%) was added to the supernatant. The reaction was carried out at room temperature and was protected from light for 16 h. Then, 0.87 mol L<sup>-1</sup> proline (Sigma, 99%) was added, and the samples were maintained at room temperature for 30 min. Toluene (Sigma) was used to extract the dansylated PAs. Aliquots (20 µL) were applied manually with a Hamilton syringe (50 µL) onto activated (1 h at 110°C, before use) glass plates (Adamant®Silica gel 60G, 0.25 mm (20 x 20cm), Macherey-Nagel) and were separated in a thin layer chromatography (TLC) developing tank, using chloroform : triethylamine (7.5:1) as a mobile phase. The plate was allowed to dry at room temperature (22 ± 2°C), and was then dried with a hair dryer to remove excess solvent. Putrescine (Put) (Sigma, 98%), spermidine (Spd) (Sigma, 99%) and spermine (Spm) (Sigma, 99%) were used as standards. The entire procedure was monitored under UV light at 254nm. Free PAs were quantified by comparison against standards by fluorescence emission spectroscopy (excitation at 350 nm and emission at 495 nm), in a Video Documentation System, using the Image Master 2.0 Software (Amersham Pharmacia Biotech 1996). The calculation of the quantitative analysis was performed based on the area obtained from the standards and the samples. The free PAs content was expressed as nmol g-1 fresh weight (FW).

# High-performance liquid chromatography (HPLC) analysis of flavonoids

Samples were extracted, filtered (Millipore 0.22 µm filter) and used for flavonoids analyses according to a previous report method (Escarpa and González, 1999). Briefly, 20 µL sample was injected into a Thermo Scientific Dionex UltiMate 3000 system (Thermo Fisher Scientific Inc., MA, USA), coupled to a quaternary pump, an Ultimate 3000RS auto sampler and a diode array detector (DAD-3000RS). Flavonoids were separated on an Ace C18 (4.6  $\times$  250 mm; 5µm) column at 25°C. Analysis was monitored at 280 nm and peak integration and calibrations were performed between 210 and 350 nm using Dionex Chromeleon software. The flow rate was 1.0 mL min<sup>-1</sup> and the mobile phase consisted of methanol (solvent A) and 0.01M phosphoric acid. The system was run with the following gradient elution program: 0-5 min, 0.5% A, 5-10 min, 50% A, 10-15 min 70% A, 15-20 min, 80% A, 25-30 min 100% A and to 30-35 min 5%.

Flavonoids were quantified by determining peak areas under the curve in the HPLC calibrated against known amounts of standards. Identification of each peak in unknown peak was confirmed by the retention time and characteristic spectra of the standards. The interassay coefficient of variation (CV) was 3.7% (n = 15) and the intra-assay CV was 4% (n = 8).

## Statistical analysis

All the results are given as means  $\pm$  standard deviation. Differences between variables were tested for significance using a one-way ANOVA procedure, followed by Tukey's test, at a significance level of p<5%.

**Table 1.** pH, soluble solids (°Brix), titratable acidity (g citric acid 100 g<sup>-1</sup>) and ratio (SS/TA of jurubeba raw and subjected to different boiling times (10, 20, 30 and 40 min) and types of preservatives (oil and vinegar).

Time (min)	Oil	Vinegar	
pH			
Raw	$5.58 \pm 0.07^{abA_*}$	$5.58 \pm 0.07^{aA}$	
10	$5.45 \pm 0.03^{bA}$	$3.94 \pm 0.02^{bB}$	
20	$5.63 \pm 0.06^{aA}$	$3.90 \pm 0.00^{bB}$	
30	$5.58 \pm 0.10^{abA}$	$3.92 \pm 0.02^{bB}$	
40	$5.52 \pm 0.10^{abA}$	$3.90 \pm 0.01^{bB}$	
TA			
Raw	0.14± 0.01 <sup>aA</sup>	0.14± 0.01 <sup>bA</sup>	
10	$0.11 \pm 0.01^{abB}$	$0.33 \pm 0.07^{aA}$	
20	$0.07 \pm 0.01^{bB}$	$0.28 \pm 0.00^{aA}$	
30	$0.06 \pm 0.00^{bB}$	0.32± 0.01 <sup>aA</sup>	
40	0.10± 0.00 <sup>abB</sup>	0.32± 0.00 <sup>aA</sup>	
SS			
Raw	$15.33 \pm 0.7^{bA}$	15.33 ± 0.7 <sup>aA</sup>	
10	$18.7 \pm 2.7^{abA}$	$9.9 \pm 0.6^{abB}$	
20	$21.3 \pm 5.1^{aA}$	$9.87 \pm 0.8^{abB}$	
30	$19.63 \pm 1.7^{abA}$	$9.03 \pm 0.3^{bB}$	
40	$18.73 \pm 3.7^{abA}$	$9.27 \pm 0.1^{bB}$	
SS/TA			
Raw	$107.95 \pm 11.59c^{A}$	107.95 ± 11.59 <sup>aA</sup>	
10	$174.00 \pm 31.43^{bA}$	$31.03 \pm 8.12^{bB}$	
20	283.61 ±31.04 <sup>aA</sup>	$35.05 \pm 2.33^{bB}$	
30	$295.08 \pm 6.07^{aA}$	$27.47 \pm 2.01^{bB}$	
40	176.59 ± 36.29 <sup>bA</sup>	$28.74 \pm 0.99^{bB}$	

<sup>\*</sup>Within columns means followed by different lowercase letters and upper case on lines are significantly different based on Tukey's ANOVA test (P < 0.05).

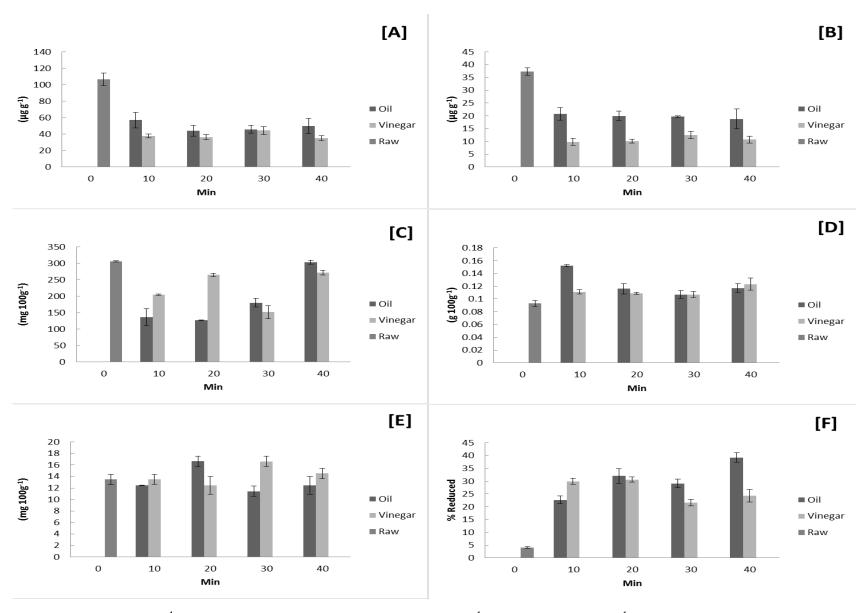
# **RESULTS AND DISCUSSION**

Cooking time influenced the quality parameters (pH, soluble solids and soluble solids/titratable acidity) in *S. paniculatum* fruit that were conserved in oil (Table 1). The results showed a slight variation in quality parameters for fruits that were pickled in vegetable oil and cooked for different lengths of time. The highest values were observed for fruits boiled for 20 min. Jurubeba fruit that were pickled in vinegar, which has a high acidity, exhibited a lower pH than those preserved in oil. Besides consumer acceptance, the pH affects many chemical processes, such as protein properties (denaturation), enzymatic activity and also the growth of microorganisms (Stippl et al., 2004).

This effect on pH was reflected in the titratable acidity of samples preserved in vinegar, as well as in oil. Fruit that were boiled for 20 min and preserved in oil showed higher levels of SS, although no difference was observed

at other cooking times. When pickled in vinegar, the values for SS after 20 min were similar to those at 10 min. The ratio of SS/TA increased with the use of oil as a preservative and with the cooking time, as compared to that of raw fruits. However, this ratio decreased proportionally with an increase in cooking time in fruits preserved in vinegar.

In the present study, chlorophyll and carotenoid levels decreased after cooking as compared to raw fruits (Figure 2A and B). The loss of green color in many vegetables after cooking is a frequent problem that affects the quality of many canned fruits and vegetables. At room temperature, chlorophylls (a and b) are stable, but when temperatures above 50°C are used, chlorophyll levels can be affected (Andrés-Bello et al., 2013). The decreased levels of chlorophyll following heat treatment is due to the conversion of chlorophyll to pheophytin, which is attributable to a change in pH during thermal processing and the hydrogen ions cause the replacement



**Figure 2.**Total chlorophyll (ug g<sup>-1</sup>) (A), total carotenoids (ug/g) (B), total flavonoids (100 mg g<sup>-1</sup>) (C), total phenols (100 g g<sup>-1</sup>) (D), ascorbic acid (mg 100g-1) (E) and (F) antioxidant capacity (TEAC mmol L<sup>-1</sup>,% reduced DPPH) (F) in "Jurubeba" raw and subjected to different boiling times (10, 20, 30 and 40 min) and types of preservatives (oil and vinegar).

of the Mg porphyrin ring (Minguez-Mosquera et al., 1989). Other studies have also described the loss of chlorophyll in broccoli by 67.87% after cooking (Pellegrini et al., 2010).

After 10 and 40 min of boiling, jurubeba preserved in vinegar contained less chlorophyll than jurubebas preserved in oil. A loss of chlorophyll was observed, regardless of the type of preservative tested. Zhang and Hamauzu (2004) observed that cooking time was a key factor in reducing the chlorophyll content in broccoli. In the present study, no difference in chlorophyll content was observed after 20 or 30 min of boiling, regardless of the type of preservative used. Thus, it is concluded that a cooking time of 20 min, which is commonly used by preserve manufacturers, is the ideal time to maintain the green color of the jurubeba fruit.

Thermal processing did not cause an increase in the level of total carotenoids in jurubeba (Figure 2B) as compared to that in raw fruits, but the type of preservative did influence carotenoid levels; fruits preserved in oil contained higher levels of carotenoids than those preserved in vinegar. Generally, due to the high temperature in the cooking process, matrix disruption occurs, promoting the extraction of compounds in the cell and many of these compounds migrate into the cooking water. De Sá and Rodriguez-Amaya (2003) suggested that an increase in carotenoid content occurs after cooking; however, studies have demonstrated lower levels of carotenoids after thermal processing (Zhang and Hamauzu, 2004), which confirm the results of this study.

After cooking, the level of total phenols increased at all cooking times as compared to that in raw fruits, regardless of the preservative used (oil or vinegar) (Figure 2C), especially when fruits were preserved in oil and boiled for 10 min. A similar increase in total phenols after heat treatment was described for eggplants (Solanum melongena) (Salerno et al., 2014).

In contrast, flavonoid levels were strongly affected by the type of preservative (Figure 2D). In vinegar, fruits showed higher levels of these polyphenols as compared to raw fruits. This increase in the content of phenols and flavonoids might be due to the high-temperature extraction/cooking time, which promoted denaturation of the fruit matrix and increased the extractability of these compounds (Turkmen et al., 2005; Zhang and Hamauzu, 2004). Cooking promotes the softening of the cell wall and other components of cells, such as vacuoles and the apoplast, releasing the phenolic compounds. Another factor that might contribute to the increase in the polyphenol content is the decomposition of phenolic compounds that are linked to fibers (cellulose and pectin) (Gökmen et al., 2009), or even the disruption of the bonds between phenols and sugars, which contributes to the increase in the polyphenol concentration (Singleton et al., 1999).

Ascorbic acid levels (Figure 2E) in fruits boiled for 20 min and preserved in oil were higher than those in raw

jurubeba or those preserved in vinegar. However, this trend disappeared when cooking time increased, and fruits preserved in vinegar showed a higher ascorbic acid content.

The antioxidant capacity (Figure 2F) of jurubeba fruit, cooked and preserved in oil, increased significantly at all cooking times. The data obtained from the analysis of antioxidants in this study showed that heat treatment increased the antioxidant capacity in jurubeba.

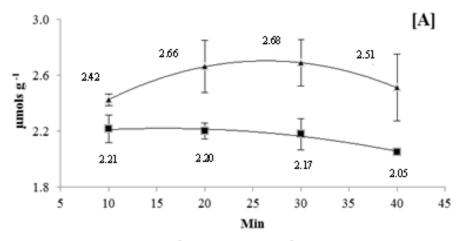
No difference in the antioxidant capacity was observed between types of preservative after cooking for 20 min, but jurubeba fruit that was preserved in oil showed a higher antioxidant capacity after 30 and 40 min of boiling. Probably this effect is attributed to the release of antioxidant compounds from the fruit matrix as a function of the increase in temperature.

There was no difference in the polyamine content of jurubeba preserved in soybean oil between different cooking times. In addition, the jurubeba preserved in vinegar showed lower values and a decrease in putrescine was observed with an increase in the cooking time. However, spermidine and spermine levels increased together with the cooking time in jurubeba preserved in vinegar and decreased in fruits preserved in oil.

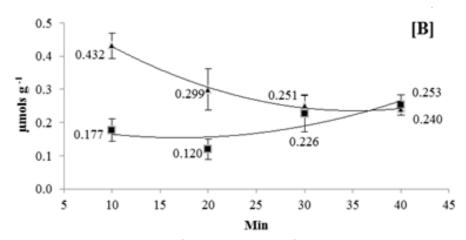
The presence of the three polyamines in the preserves was expected, because they occur naturally in fruits and vegetables (Figure 3). As compared to the levels of polyamines in raw jurubeba (putrescine, 2.01  $\mu mols~g^{-1}$ , spermidine 0.10  $\mu mols~g^{-1}$  and spermine 1.77  $\mu mols~g^{-1}$ ) and after thermal processing, there was an increase in the levels of putrescine and spermidine regardless of the cooking time, whereas spermine levels decreased in fruits stored in vinegar and increased in those preserved in oil.

Some studies have demonstrated that the cooking process can induce changes in the levels of polyamines. Rossetto et al. (2015) observed a reduction in the content of putrescine, spermine and spermidine in vegetables such as carrots, broccoli, cabbage and beetroot cooked in water. However, in others studies, the cooking process did not alter the levels of these amines (Eliassen et al., 2002). The cooking of jurubeba altered the levels of putrescine, spermidine and spermine. It has been reported that polyamines can be leached in boiling water, leading to a decrease in the levels of polyamines after the cooking of some vegetables. Some studies showed that a putrescine loss of about 20-25% occurred in broccoli and celery and of 40% in cauliflower and asparagus. Similarly, a 10-20% loss of spermidine occurred in broccoli and celery, and a decrease of 20-30% in cauliflower and asparagus (Ziegler et al., 1994).

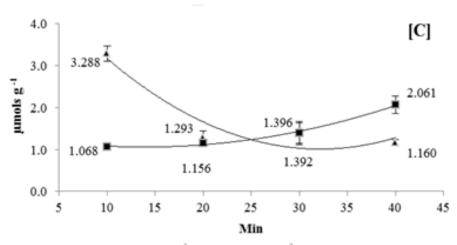
Our results show that in addition to changes in the levels of polyamines induced by cooking in water, the type of preservative also has an effect. Jurubeba preserved in vinegar contained lower levels of putrescine, ranging from 2.05 to 2.21 µmol/g, where as in those preserved in oil, the levels ranged between 2.42 and 2.68



- Oil Putrescine =  $0.001x^2 0.0546x + 1.9845$  R<sup>2</sup> = 0.99
- Vinegar: Putrescine =  $0.0003x^2 0.0087x + 2.1527 R^2 = 0.97$



- $\triangle$  01 spermine = 0.0003x<sup>2</sup> 0.0215x + 0.6134 R<sup>2</sup> = 0.99
- Vinegar: Spermine =  $0.0002x^2 0.0072 + 0.2154 R^2 = 0.71$



- Oil spermine =  $0.0044 \,\mathrm{m}^2 0.2826 + 5.5517 \,\mathrm{R}^2 = 0.90$
- Vinegar: Spermine = 0.0015x<sup>2</sup> 0.0404 + 13406 R<sup>2</sup> = 0.99

**Figure 3.** Putrescine (A), spermidine(B) and spermine ( $\mu$ mols  $g^{-1}$ ) (C) ( $\mu$ mols  $g^{-1}$ ) in jurubeba fruits raw and subjected to different boiling times (10, 20, 30 and 40 min) and types of preservatives (oil and vinegar).

Table 2. Polyphenols content (isoorientine	, rutine and acid	d caffeic) of rav	w jurubeba fruits	and
subjected to different boiling times (10, 20	, 30 and 40 min	) and types of	preservatives (oil	and
vinegar).				

Preservative	Time	Polyphenols (mg 100g <sup>-1</sup> )		
	(min)	Isoorientin	Rutin	Caffeic acid
Oil	Raw	21.3 ± 2.16 <sup>b</sup> *	$47.90 \pm 6.88^{\circ}$	$0.10 \pm 0.02^{h}$
	10	$81.6 \pm 8.67^{a}$	$75.38 \pm 6.14^{a}$	$0.41 \pm 0.07^{f}$
	20	$80.4 \pm 7.81^{a}$	$69.38 \pm 5.95^{b}$	$0.35 \pm 0.04^{g}$
	30	$83.7 \pm 11.70^{a}$	$80.94 \pm 5.80^{a}$	$0.40 \pm 0.05^{e}$
	40	83.0 ± 9.91 <sup>a</sup>	76.45 ± 7.47 <sup>a</sup>	$0.45 \pm 0.07^{d}$
Vinegar	10	83.0 ± 17.04 <sup>a</sup>	68.47 ± 4.41 <sup>a</sup>	$0.56 \pm 0.10^{c}$
	20	$85.6 \pm 3.03^{a}$	$59.59 \pm 4.25$	$0.66 \pm 0.01^{b}$
	30	$81.0 \pm 0.79^{a}$	$55.67 \pm 4.42^{b}$	$0.64 \pm 0.06^{b}$
	40	82.2 ± 4.85 <sup>a</sup>	61.36 ± 3.23 <sup>b</sup>	$0.76 \pm 0.09^{a}$

<sup>\*</sup>Within columns means followed by different lowercase letters are significantly different based on Tukey's ANOVA test (P < 0.05).

µmol/g. In vinegar preserves, lower putrescine content might indicate a better fruit quality.

Biogenic amines, especially histamine, putrescine and cadaverine have been suggested to be indicators of the deterioration of some types of food such as fresh fish, meat and vegetables (Riebroy et al., 2004). These amines are important for nutrition and health. Spermidine and spermine are directly related to DNA and to cell division and their levels in jurubeba fruits cooked for 20 min were lower than at other cooking times. These results might be relevant to individuals with certain neoplasias, who posses low levels of these compounds, especially putrescine. However, spermine is important for the regulation of nitric oxide levels and absorption and can contribute to the balance of excessive production of nitric oxide. This free radical (NO) is linked to tumor progression (Til et al., 1997) and spermine has also been linked with a decrease in inflammation (Moinard et al., 2005).

Polyamines occur naturally in plants. The levels of polyamines found in this study are important in relation to the consumption of these substances, because they may be related to some heart diseases and some types of cancer. Polyamines do not cause cancer, but accelerate tumor growth. Increased levels due to the synthesis of polyamines in animal tissues and to food intake can cause increased cell growth (Mandalet al., 2015).

From the analysis of polyphenols via HPLC, isoorientin, rutin and caffeic acid were identified. All polyphenols were found at a lower concentration in raw Jurubeba fruits, but increased when the fruits were cooked, but the difference was not significant (Table 2). It was observed that a 10 min cooking time is sufficient to reach maximum polyphenol content. However, in other analyses, it was found that 20 min is the optimal time. A cooking time of 20 min can be used without altering the polyphenol

content. Jurubeba fruits preserved in oil had a higher content of rutin, whereas those stored in vinegar had higher levels of caffeic acid. This increase in the release of flavonoids (rutin and isoorientin) and phenolic acid (caffeic acid) suggests that the antioxidant capacity within jurubeba fruits might increase or remain unchanged after cooking and when preserved either in oil or vinegar. Phenolic compounds are also water-soluble, rendering them susceptible to leaching. In our study, this effect was not observed. The cooking or the canning process did not influence the content of isoorientin, rutin or caffeic acid. Furthermore, it has been described that a decrease in these compounds occurs due to leaching into the brine rather than via oxidation (Chaovanalikit and Wrolstad, 2004).

From the obtained data, the treatment using 20 min of cooking appears to give the best results. In addition to being the most widely used treatment in homemade jurubeba preserves, this treatment did not induce changes in chlorophyll levels, an important parameter for visual analysis. In addition, some quality parameters such as pH and SS were high following this length of cooking time, as well as in both types of preservatives used, which was reflected in the ratio between SS and TA. Following cooking for 20 min, no change in the level of carotenoids were observed that could decrease the quality of the jurubeba fruits. No changes were also observed in the total phenol content, antioxidant capacity or the contents of isoorientin, rutin and caffeic acid. A further analysis confirms a cooking time of 20 min to be optimal for the lower contents of spermidine and spermine. Using this thermal treatment and vinegar as a preservative, the fruits had the lowest level of putrescine. Jurubeba preserved in oil retained more carotenoids, and a higher content of putrescine, spermidine and antioxidant capacity, and showed the highest values of pH, SS and

SS/TA.

#### Conflict of interest

The authors declare that they have no conflict of interest.

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## **Abbreviations**

**Cla**, Chlorophyll *a*; **Clb**, chlorophyll *b*; **Pas**, polyamines; **Put**, putrescine; **Spd**, spermidine; **Spm**, spermine; **TA**, titratable acidity; **SS**, soluble solids.

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