

Quality attributes of nance (*Byrsonima crassifolia*) fruits as affected by storage temperature and maturity at harvest

¹Rivera-Correa, K. A., ^{1*}Valle-Guadarrama, S., ²Alia-Tejacal, I.,
³Arevalo-Galarza, M. L. C., ¹Pérez-López, A. and ⁴Guerra-Ramírez, D.

¹Departamento de Ingeniería Agroindustrial, Universidad Autónoma Chapingo, Mexico-Texcoco km 38.5, Chapingo, 56230, Texcoco de Mora, Mexico

²Facultad de Ciencias Agropecuarias, Universidad Autónoma del Estado de Morelos, Av. Universidad No. 1001, Chamilpa, Cuernavaca, 62209, Morelos, Mexico

³Instituto de Recursos Genéticos y Productividad, Colegio de Postgraduados, Mexico-Texcoco km 36.5, Montecillo, 56230, Texcoco de Mora, Mexico

⁴Laboratorio de Productos Naturales, Departamento de Preparatoria Agrícola, Universidad Autónoma Chapingo, Mexico-Texcoco km 38.5, Chapingo, 56230, Texcoco de Mora, Mexico

Article history

Received: 2 August 2020

Received in revised form:

21 May 2021

Accepted:

9 July 2021

Abstract

Nance (*Byrsonima crassifolia*) fruit is harvested when natural abscission from the plant occurs. At this stage, the shelf life is less than 5 d in ambient conditions. The aim of the present work was thus to determine how quality attributes of nance fruits are modified as a function of ripening on the tree, physiological condition at harvest, and storage temperature. Fruits at three maturity stages (green, transient, and yellow) were harvested and stored at 15 and 25°C. As fruits ripened, the hue angle turned to yellow, and lightness and chroma increased, but carotenoid content decreased. The contents of total soluble solids, total sugars, and reducing sugars increased; however, the total soluble phenols, flavonoids, and antioxidant activity decreased. It was possible to harvest at a physiological stage previous to abscission maturity even though a non-climacteric pattern was identified. Handling of transient nance fruits at 15°C extended shelf life for more than 15 d, with adequate physical and compositional attributes including high concentration of bioactive compounds and antioxidant activity. Content of total soluble solids was identified as an attribute suitable for developing a harvest index for nance fruits.

© All Rights Reserved

Keywords

Byrsonima crassifolia,
maturity at harvest,
quality attributes,
refrigeration,
ripening

Introduction

Byrsonima crassifolia (L.) Kunth grows in tropical and subtropical areas of the United States (Florida), Mexico, Central America, Brazil, and other South American countries. Several studies have highlighted its importance because the leaves, fruits, seeds, barks, and branches contain bioactive compounds that are anti-inflammatory (Pérez and Muñiz, 2016), antidepressant (Herrera-Ruiz *et al.*, 2011), antidiabetic (Pérez and Muñiz, 2016), and have antioxidant properties that reduce free radicals (Mariutti *et al.*, 2014).

Nance fruit is a rounded drupe, with diameter ranging from 0.9 (da Silva *et al.*, 2016) to 3.0 cm (Duarte and Paull, 2015), and a seed that is 10.2 - 14.4% of its total weight (Duarte, 2011). Its yellowish pulp and skin change from green to yellow-orange or red upon ripening (Neves *et al.*, 2015; Rivas-Castro *et al.*, 2019). The shelf life of this

fruit is less than 5 d at 20°C (Rivas-Castro *et al.*, 2019), thus limiting its commercialisation to local markets and production areas. Due to its nutraceutical properties, several phytochemical components (Séfora *et al.*, 2019) have been extracted for alternative uses. Whether consumed fresh or processed to extract bioactive compounds, postharvest life must be extended to preserve quality.

Nance fruit is collected at the time of natural abscission (Medina-Torres *et al.*, 2004), but it can remain attached to the plant long after ripening has elapsed (Medina-Torres *et al.*, 2012; da Silva *et al.*, 2016), thus suggesting that it is consumed under senescent conditions. The harvest index of nance fruit has not been well determined since its ripening has not been fully characterised. Based on flower marking and development monitoring, da Silva *et al.* (2016) identified some of the changes that occur during ripening, such as a decrease in starch content and an increase in sugars and total soluble solids.

*Corresponding author.
Email: svalleg@taurus.chapingo.mx

Rivas-Castro *et al.* (2019) suggested that the fruit can be harvested at a stage prior to natural abscission, which can lengthen its shelf life. To confirm these ideas, it is necessary, first, to characterise the ripening process of fruits when they are still attached to the tree, and then evaluate their physiological behaviour during the postharvest period at different ripening stages. The aim of the present work therefore was to assess the changes in quality attributes of nance fruits as a function of ripening on the tree, physiological condition at harvest, and storage temperature.

Materials and methods

Plant material

Fully developed nance fruits were harvested in Amacuzac, Morelos, Mexico (18° 34' 29" N; 99° 20' 46" W; 900 m altitude) at three stages of maturity. Based on experience of producers, green (G) was associated with physiological maturity, yellow (Y) with abscission maturity, and transient (T) yellow-green with an intermediate stage between the two conditions. Damage-free fruits were selected as experimental material.

Experimental organisation

Batches of 100 g of G, T, and Y fruits constituted experimental units (Eu). Half of the batches were stored at 15 (± 2)°C and 80 (± 3)% relative humidity (Hr), and half at 25 (± 1)°C and 70 (± 3)% Hr. From this, treatments G₁₅, T₁₅, Y₁₅, G₂₅, T₂₅, and Y₂₅ were formed. Throughout a 21-d storage, Eu were periodically removed from treatments to measure epicarp colour, respiration, ethylene production, firmness, acidity, and contents of total soluble solids, total sugars, reducing sugars, vitamin C, carotenoids, total soluble phenols, flavonoids, and antioxidant activity. Data recorded on the 1st d of sampling were used as the baseline to evaluate changes during ripening on plant. In addition, postharvest behaviour was monitored considering the maturity at harvest, temperature, and storage time as variation factors. Values measured on Y fruits at harvest, identified with a subscript R, were taken as reference to evaluate postharvest behaviour.

Response variables

Colour was measured on five fruits with a Hunter Lab colorimeter (Hunter Lab®, USA). Results were averaged in each Eu, and expressed as lightness (L*), hue angle (H*), and chroma (C*) (McGuire, 1992). Respiration and ethylene

production rates were evaluated following the procedure described by Mahajan *et al.* (2014) with modifications. Experimental units were placed in 475-mL containers for 1 h; the headspace was analysed in terms of CO₂ and C₂H₄ concentrations with a gas chromatograph (Agilent Technologies® 7890A, USA), operated at 150, 80, and 170°C, in injector, oven, and detectors, respectively, using nitrogen as carrier gas. Based on the changes in concentration, the routine time, fruit mass, free container volume, respiration rate, and ethylene production rate were evaluated in mL/kg h and µL/kg h, respectively, using Eq. 1.

$$g_{CO_2} = \frac{\Delta y_{CO_2} V_c}{m t_r}; \quad g_{C_2H_4} = \frac{\Delta y_{C_2H_4} V_c}{m t_r} \quad (\text{Eq. 1})$$

Firmness was measured with a texture analyser (TA-TX2i, Stable Micro Systems, UK) using a 5-mm spherical probe that compressed fruits up to 5 mm at 2-mm/s speed. Results of five fruits were averaged and expressed in Newton (N). Total soluble solids (TSS) content, expressed as °Brix, was determined in juice extracted from fruits with a refractometer (CVQ-4012 VelaQuin®, Mexico). Titratable acidity (TA), expressed as percentage of citric acid, was also determined in juice of fruits following method 942.15 of the AOAC (1990), through titration with 0.1 N NaOH. Total sugars were quantified with the phenol-sulphuric method (Safářík and Šantrůčková, 1992) with modifications; fruit pulp was diluted (1:200) in distilled water, 1 mL of such dilution was mixed with 0.6 mL of a 5% phenol solution, and 3.6 mL of concentrate sulphuric acid were then quickly added. The mixture was agitated in vortex for 10 s, allowed to stand for 1 h, and subjected to absorbance evaluation at 485 nm with a DR 500 UV-vis HACH spectrophotometer (Germany), and a phenol-sulphuric mixture without sample were used as blank. The quantification of total sugars was aided by a standard glucose curve prepared in the range of 0 to 100 µg/mL. The DNS (3,5-dinitrosalicylic acid) method (Miller, 1959) was used to determine the reducing sugar content. A working solution (w/v) of 1.6% NaOH, 30.0% tetra hydrated sodium potassium tartrate, and 1.0% DNS was prepared. Fruit pulp was diluted (1:10) in distilled water, and 1 mL of this dilution was mixed with 1.0 mL of the working solution; the mixture was boiled for 5 min and cooled in an ice-water bath. Then, 10 mL of distilled water were added, and the mixture was shaken and allowed to stand for 15 min. Absorbance was measured at 540 nm using a DR

500 UV-vis HACH spectrophotometer (Germany). A similar procedure was applied to a blank of distilled water instead of juice. Quantification of reducing sugars was aided with a standard curve of glucose prepared in the range of 0 to 2.0 mg/mL.

To evaluate the contents of total soluble phenol (TSP) and total flavonoid (TFC), and antioxidant activity, extracts were prepared following the procedures described by Dudonné *et al.* (2009) and Miranda-Molina *et al.* (2019) with some modifications: 1.0 g of fruits pulp was mixed with 10 mL of 80% methanol, and stirred in vortex (Symphony VWR, China) for 1 min at 1,500 rpm; pH was adjusted to 4.0, and each mixture was again stirred at 1,500 rpm for 3 min; then sonication in a Cole-Parmer bath (Cole-Parmer 8890, USA) was applied for 30 min in the dark, and agitated in incubator (INO-650M, Prendo®, México) at 150 rpm and 30°C for 30 min. The mixture was then centrifuged (J-600 SolBat, Mexico) at 4,000 rpm for 14 min. Supernatants were recovered, and volumes were adjusted to 10 mL with 80% methanol. The procedure was carried out at 25°C.

The TSP was determined with the method of Singleton and Rossi (1965), following a procedure adapted to microplates (Cao *et al.*, 2020) using a Synergy™ HTX spectrophotometer (BioTek Instruments, Inc., USA). Samples of 25 µL of extract were mixed with 20 µL of the Folin-Ciocalteu reagent, which was diluted at a 1:1 ratio of the commercial 2 N solution (Sigma-Aldrich, Co., Germany) with distilled water. The mixture was neutralised with 30 µL of 20% Na₂CO₃ (w/v) and 125 µL of distilled water. The treatments were placed in the dark for 30 min, and absorbance was read at 760 nm. TSP quantification was based on a standard curve of gallic acid (0 - 90 mg/mL), and the results were expressed in mg equivalents of gallic acid per 100 g of fresh material (mg/100 g).

The aluminium chloride colorimetric method described by Chang *et al.* (2002) was used with modifications to evaluate TFC. A dilution was made with 1 mL of the extract and 1 mL of distilled water. Subsequently, 0.5 mL of this dilution was dissolved in 2.5 mL of distilled water. Then, 0.15 mL of 5% NaNO₂ was added, and the sample was left to stand for 5 min in the dark. Next, 0.3 mL of 10% AlCl₃ was added, and the mixture was again left to stand for 5 min in the dark. Next, 1 mL of 4% NaOH was added and stirred at 3,000 rpm for 3 min. Finally, aliquots of 200 µL were subjected to absorbance measurement at 510 nm with a Synergy™ HTX spectrophotometer, and TFC was determined using a standard curve of catechin

prepared in the range of 0 to 290 µg/mL. Results were expressed in mg equivalents of catechin per 100 g of sample (mg/100 g).

To evaluate total carotenoids, an extract was prepared by blending fruit pulp and a solvent of hexane:acetone:ethanol (2:1:1) in a 1:5 ratio and leaving it to stand for 1 h in the dark (Speek *et al.*, 1988). Carotenoids were quantified by absorbance measurements at 450 nm and a standard curve of β-carotene (60 - 300 µg/L).

The method of Nielsen (2010) was applied to evaluate vitamin C (Vc) content. Fruit pulp was diluted (1:2) in distilled water and titration was performed with a 2,6-dichlorophenolindophenol solution at 0.02%. Quantification was aided by a standard curve of Vc prepared in the range of 0 to 250 mg/L.

The antioxidant activity (AA) was determined with the FRAP (ferric reducing antioxidant power) (Benzie and Strain, 1996) and ABTS (2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid)) (Re *et al.*, 1999) assays. For the FRAP assay, a buffer solution of 300 mM sodium acetate trihydrate with pH 3.6 was prepared by mixing 20 mM ferric chloride hexahydrate and 2,4,6,- 10 mM Tris (2-pyridyl)-s-triazine (TPTZ) dissolved in 40 mM HCl. The FRAP solution (180 µL) was added directly to samples (20 µL), and absorbance was immediately measured at 530 nm with a Synergy™ spectrophotometer. The ABTS assay was based on a 7.4 mM solution of ABTS dissolved in a 2.6 mM sodium persulfate solution, which was left to react for 16 h in darkness before use. Next, 20 µL of extract were mixed with 230 µL of the ABTS solution. The decrease in absorbance at 734 nm was measured after 10 min with a Synergy™ spectrophotometer. Results were expressed, in both cases, as mg equivalent of Trolox per 100 g of fresh sample (mg/100 g).

Data analysis

The experimental organisation considered temperature (15 and 25°C), maturity at harvest (green, transient, and yellow), and storage time as variation factors. Data were subjected to analyses of variance together with routines focused on the comparison of treatment means using the Tukey test at a significance level of 0.05. All evaluations were made in triplicate.

Results and discussion

Colour

On the day of harvest, there was significant

Table 1. Quality attributes of nance fruits harvested at different stages of maturity.

Variable	Maturity			HSD
	Green	Transient	Yellow	
Firmness (N)	55.22 ^a (0.46)	51.75 ^b (0.53)	21.04 ^c (0.72)	2.13
Hue angle (degrees)	100.89 ^a (0.23)	89.23 ^b (0.29)	78.99 ^c (0.48)	1.27
Lightness (%)	60.57 ^c (0.48)	71.52 ^b (0.25)	75.36 ^a (0.78)	2.01
Chroma	40.64 ^c (0.48)	51.39 ^b (0.21)	61.10 ^a (1.23)	2.84
TSS (°Brix)	13.34 ^c (0.17)	17.00 ^b (0.42)	21.31 ^a (0.61)	1.62
Acidity (%)	0.23 ^a (0.01)	0.23 ^a (0.01)	0.21 ^a (0.01)	0.029
Total sugar (%)	11.47 ^b (0.57)	16.9 ^a (0.69)	19.49 ^a (1.27)	3.29
Reducing sugar (%)	7.69 ^c (0.2)	10.39 ^b (0.12)	13.08 ^a (0.32)	0.83
Vitamin C (mg/100 g)	9.83 ^c (1.38)	49.65 ^a (3.37)	24.98 ^b (2.77)	9.69
Carotenoid (µg/100 g)	64.35 ^a (5.42)	34.35 ^b (1.98)	26.46 ^b (0.93)	12.40
TSP (mg/100 g)	938.05 ^a (54.51)	581.47 ^b (11.89)	267.14 ^c (8.68)	119.74
Flavonoid (mg/100 g)	73.90 ^a (6.25)	60.44 ^b (1.8)	16.34 ^c (1.03)	13.36
AA (ABTS) (mg/100 g)	2151.22 ^a (107.79)	1849.1 ^a (104.65)	547.5 ^b (31.83)	325.70
AA (FRAP) (mg/100 g)	71.47 ^a (2.52)	61.72 ^b (1.88)	22.78 ^c (0.90)	6.93

TSS: total soluble solid; TSP: total soluble phenol; AA: antioxidant activity. Different lowercase superscripts indicate significant difference. HSD: honest significant difference (Tukey, 0.05). Values in parentheses are standard errors.

difference in hue angle (H^*) ($p \leq 0.05$) among maturity stages (Table 1), with values between 78.0 and 80.7° in Y fruits, 88.2 and 90.3° in T fruits, and between 100.1 and 101.6° in G fruits. Lightness (L^*) varied at harvest between 72.0 and 77.0% in Y fruits, 70.4 and 72.1% in T fruits, and between 58.9 and 62.2% in G fruits, with significant difference ($p \leq 0.05$) between them (Table 1). Chroma was also significantly different ($p \leq 0.05$) among fruits harvested at different maturity stages (Table 1), with values of 58.2 to 66.8 in Y fruits, 50.5 to 51.6 in T fruits, and 38.6 to 41.9 in G fruits. Fruit ripening is commonly characterised by epicarp colour modification (Cherian *et al.*, 2014). In fact, tonality was the criterion used to classify the physiological condition of fruits as green (G), transient (T), or yellow (Y) in condition of abscission. This means that ripening on the plant was characterised by hue

angle modification from green to yellow, which is a typical feature of the studied nance fruits, accompanied by the increase in lightness and chroma.

To assess the behaviour of fruits after harvest, the colour attributes of fruits at abscission maturity (Y fruits) were used as reference (R), with average values of 79.0° (H^*_R), 75.36% (L^*_R), and 61.1 (C^*_R). During storage, Y fruits showed a decrease in H^* to 65.9-69.1° and values were not statistically different ($p > 0.05$) (Figure 1B), thus indicating that colour change was not interrupted, although the change was a transition from yellow to brown. G_{15} fruits did not reach H^*_R during storage, and maintained H^* equal to 85.8 (± 1.4)° after 21 d. However, G_{25} fruits reached H^*_R in about 10 d, with a sudden change between 9 and 12 d; appearance deteriorated significantly from day 9,

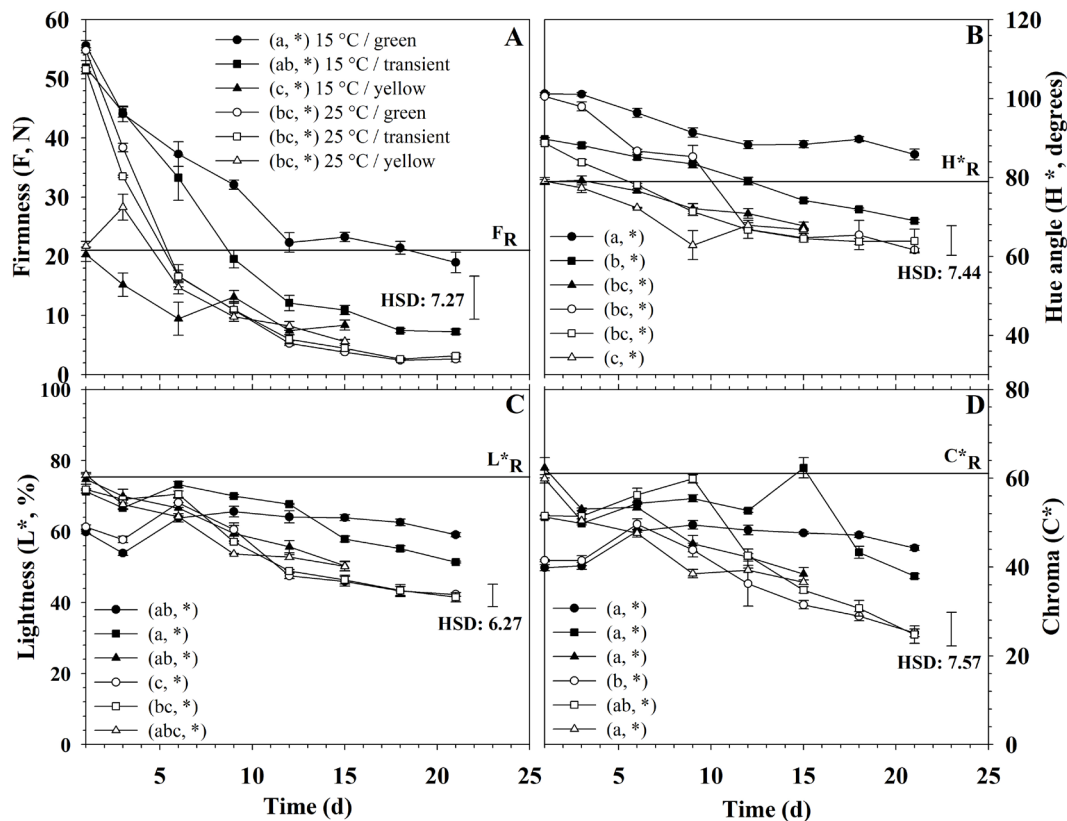


Figure 1. Firmness and colour attributes of nance fruits postharvest. Asterisk (*) indicates significant modification over time. Similar lowercase letters indicate no significant difference ($p > 0.05$) between batches. HSD: honest significant difference. Error bars indicate standard errors.

when H^* was $61.6 (\pm 0.5)^\circ$. At the end of storage, there was no transition to yellow tones, but rather to a brown hue. T_{15} and T_{25} fruits reached H^*_R in 12 and 6 d, respectively, with gradual modification, reaching values of $69.0 (\pm 0.4)^\circ$ and $63.9 (\pm 3.0)^\circ$, which were appreciated as transitions from green-yellow to yellow and later to brown. In the case of L^* , a decrease was observed in all the fruits during storage as of day 6, and fruits harvested at green or transient stages did not reach L^*_R (Figure 1C).

Storage temperature did not affect ($p > 0.05$) Y fruits, which achieved values between 47.7 and 49.0% after 15 d. In contrast, storage temperature affected G and T fruits: G_{15} and T_{15} fruits lost less lightness than G_{25} and T_{25} fruits ($p \leq 0.05$). After 21 d of storage, the highest lightness was found in G_{15} , followed by T_{15} , and the lowest in G_{25} and T_{25} , which were statistically equal. Yellow nance fruits have been reported to have 70.25% lightness and 87.52° hue angle (Moo-Huchin *et al.*, 2014), which were similar to those of ripe yellow fruits in the present work.

Regarding C^* , once the fruits were harvested, there was a significant decrease

($p \leq 0.05$). This attribute was temperature-dependent ($p \leq 0.05$) in G fruits, but it was temperature-independent ($p > 0.05$) in T and Y fruits (Figure 1D). At the end of storage, the highest chroma values were found in G_{15} and T_{15} , and the lowest in G_{25} and T_{25} , although values deviated from the reference value (C^*_R) in all the cases.

Firmness

Fruits at abscission maturity had initial firmness between 20.3 and 21.7 N, which contrasted ($p \leq 0.05$) with those of green and transient stages, with values between 51.6 and 55.6 N (Figure 1A). Softening occurs through pectolytic enzymes that degrade cell wall polymers (Wang *et al.*, 2018), and constitutes a physiological process that is normally associated with ripening. This occurred in the fruits assessed in the present work, and if fruits in green or transient stages remained attached to the tree, they would soften and reach the firmness of yellow fruits. Firmness decreased in all fruits during the postharvest period. In Y fruits, firmness was between 5.6 and 8.4 N after 15 d, when fungal development limited handling, and changes corresponded to transition to senescence since

ripening had already elapsed. The average firmness of Y fruits immediately after harvest was 21.04 N (Table 1) and used as reference (F_R) to evaluate postharvest behaviour of G and T fruits. The slowest change occurred in G_{15} , where values near F_R were observed from day 12, and fruits maintained a similar condition until the end of storage. F_R was reached in T_{15} in lesser time, between 8 and 9 d (Figure 1A), which was an expected result since ripening has progressed. After 21 d, firmness was around 7.3 N. In contrast, G_{25} and T_{25} fruits softened quickly, reaching F_R between 5 and 6 d, with values between 2.6 and 3.2 N after 21 d, thus indicating a significant effect of temperature.

Respiration and ethylene production

Respiration was significantly affected by temperature, and no effect of ripening stage was observed. From the beginning of storage, two different ($p \leq 0.05$) internally uniform ($p > 0.05$) groups were formed. One group stored at 15°C, whose respiration was $1.92 (\pm 0.09)$ mL/kg h and the other stored at 25°C, whose respiration value was $4.02 (\pm 0.14)$ mL/kg h (Figure 2A). Respiratory activity had the same order of magnitude as that reported for yellow nance by Rivas-Castro *et al.* (2019), between 7.7 and 17.3 mg/kg h, and by Neves *et al.* (2015), around 6 mg/kg h. Data suggested that nance fruit has low respiratory activity (< 10 mg/kg h; Kader, 2002). The relationship between respiration and temperature can be expressed through the Q_{10} factor (Eq. 2) (Nunes and Emond, 2003), and for every 10°C of increase in temperature, the respiratory activity of nance fruits rises 2.09 times.

$$Q_{10} = \left(R_{T_{high}} / R_{T_{low}} \right)^{\left(\frac{10}{T_{high} - T_{low}} \right)} \quad (\text{Eq. 2})$$

Throughout storage at 15°C, respiration remained without significant variations ($p > 0.05$), but at 25°C, an upward trend was observed (Figure 2A). Proliferation of fungi limited the shelf life. There was greater fungal growth at 25°C, and the highest respiratory activity occurred between 15 and 21 d, when differences, relative to the beginning of storage, became significant.

Based on respiration and firmness, Rivas-Castro *et al.* (2019) reported climacteric behaviour in nance fruits collected at transient maturity in Tabasco, Mexico. da Silva *et al.* (2016) reported similar observations in fruits of Roraima, Brazil. Both studies showed temporary increases in

CO₂ around 2 - 3 d after harvest. However, Neves *et al.* (2015) reported that nance fruits collected in Roraima, Brazil, showed non-climacteric behaviour. Softening normally accompanies ripening of nance fruits (Rivas-Castro *et al.*, 2019). In the present work, G and T fruits quickly softened during the first 9 d after harvest, and a temporary increase in respiration was expected during this period. However, such behaviour did not occur, and climacteric behaviour was not confirmed. Regarding ethylene, G and T fruits emitted $55.52 (\pm 0.97)$ and $67.50 (\pm 1.80)$ µL/kg h at 15 and 25°C, respectively, at the beginning. However, there was no effect due to maturity (Figure 2B). The cases of Y_{15} , Y_{25} , T_{15} , and G_{15} showed no significant variations ($p > 0.05$) in ethylene production during storage. Between 15 and 21 d, however, there were significant increments, concomitant with respiration in T_{25} and G_{25} . Rivas-Castro *et al.* (2019) reported temporary increases in ethylene production, but such behaviour was not verified in the present work.

Total soluble solids, acidity, and sugars

Y fruits (reference fruits) showed the highest initial content of total soluble solids (TSS), with values equal to $21.31 (\pm 0.61)$ °Brix, followed by T fruits with $17.00 (\pm 0.42)$ °Brix, and finally by G fruits with $13.34 (\pm 0.17)$ °Brix. Differences were significant among maturity stages ($p \leq 0.05$), but there was no effect of temperature ($p > 0.5$) (Figure 2C). These results indicated that TSS increased with ripening on tree. However, after harvest, TSS did not vary significantly ($p > 0.05$) in any of the batches.

In the case of titratable acidity (TA), no significant difference ($p > 0.05$) was observed at the beginning of the study among maturity stages. Values were $0.228 (\pm 0.004)\%$ in G, $0.231 (\pm 0.008)\%$ in T fruits, and $0.211 (\pm 0.010)\%$ in Y fruits. In addition, TA increased in Y fruits and in those of T_{25} , reaching values close to 0.5% in the abscission condition, and between 0.3 and 0.4% in transient fruits. In T_{15} fruits and in G fruits, TA remained without significant modification (Figure 2D). Although acidity was reported as citric acid content, it may be affected by an alteration in respiration by microbial growth. Fungal development was greater on Y fruits during postharvest, causing a gradual increase in respiration, and at tissue level, a higher concentration of CO₂ caused a decrease in pH (Valle-Guadarrama *et al.*, 2013) due to absorption as carbonic acid. Moo-Huchin *et al.* (2014) reported acidity of 0.47%, similar to values found in the present work. Also, nance acidity was similar to that

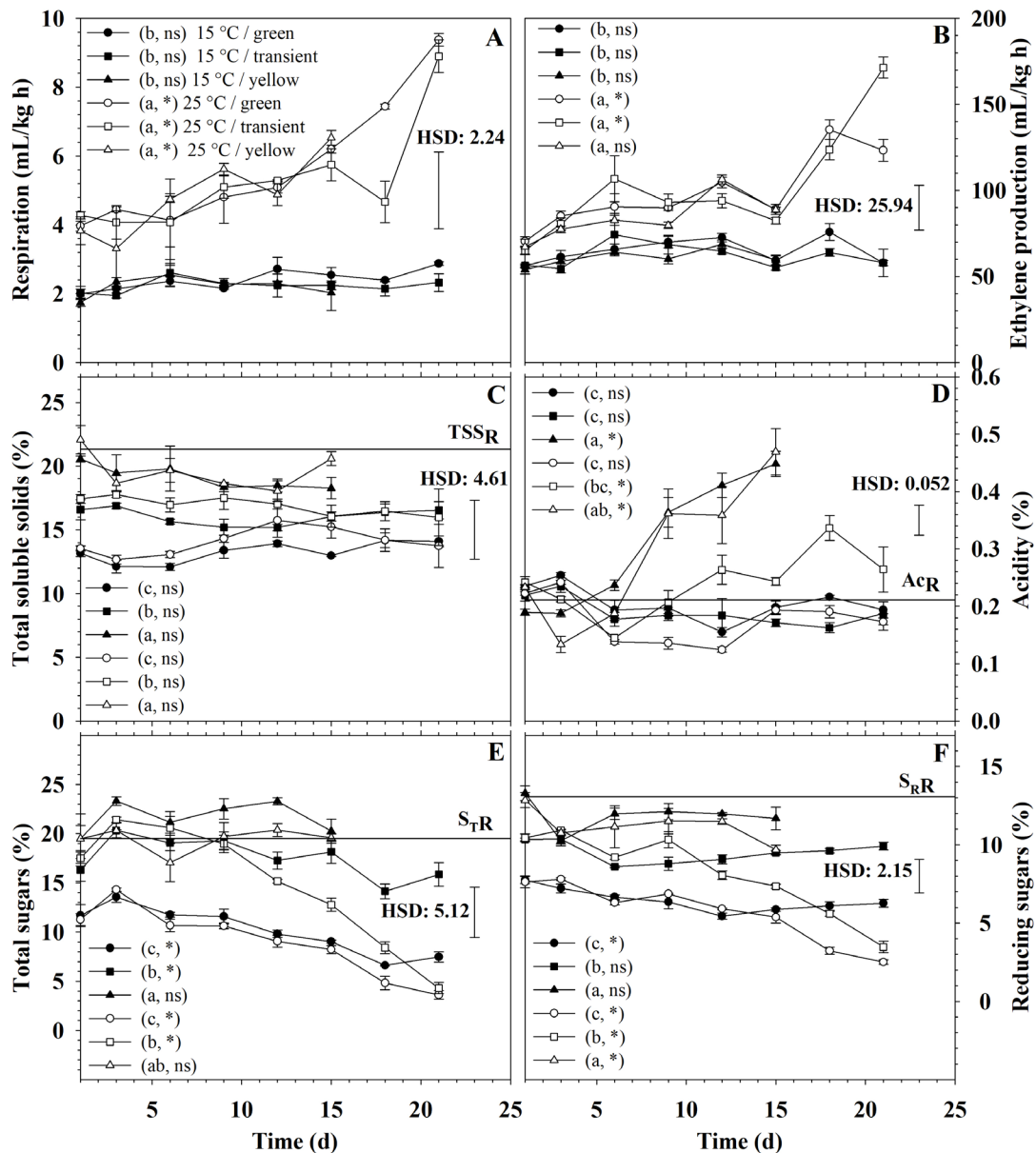


Figure 2. Respiratory activity, ethylene production, acidity, and contents of total soluble solids, total sugars, and reducing sugars of nance fruits postharvest. Asterisk (*) indicates significant modification over time. Similar lowercase letters indicate no significant difference ($p > 0.05$) between batches. HSD: honest significant difference. Error bars indicate standard errors.

of apple, which has been reported between 0.26 and 0.51% (Huang *et al.*, 2018).

Based on Figures 2C and 2D, TSS/TA ratio had initial values of $56.08 (\pm 1.73)$ in G_{15} , $60.88 (\pm 1.29)$ in G_{25} , $76.28 (\pm 6.79)$ in T_{15} , $72.12 (\pm 2.83)$ in T_{25} , $108.61 (\pm 1.94)$ in Y_{15} , and $94.52 (\pm 3.13)$ in Y_{25} . There was no significant effect of temperature, but there was an effect of maturity stage. Medina-Torres *et al.* (2004) proposed that fruits are sour if TSS/TA is between 5.1 and 8.0, bittersweet when it is 8.1 to 10, and sweet if it is above 10.0. The fruits assessed in the present work can thus be classified as sweet. Although the harvest index of nance fruits has not been well determined, Medina-Torres *et al.* (2004)

suggested the TSS/TA ratio as the best quality index for nance fruits. However, TA was not affected when fruits ripened on the tree, but TSS clearly increased, and after harvest, remained without significant variation in all the batches. For this reason, TSS could be a better variable for establishing a harvest index. On the other hand, batches behaved with different trends during storage. Y fruits experienced a decrease in TSS/TA to values between 41.0 and 44.6, while transient stage fruits remained without significant changes, and G_{15} and G_{25} fruits exhibited temporary increase on day 12, with average maximum of 90.3 and 126.6, respectively, which corresponded to a decrease in acidity in the same

period. Further investigation is needed to clarify this phenomenon.

Total sugars (S_T) and reducing sugars (S_R) had initial values (%) of 11.47 and 7.69 in G, 16.90 and 10.39 in T fruits, and 19.49 and 13.08 in Y fruits (Table 1), which indicated that S_T and S_R increased with ripening on the tree. During storage, the sugar content of Y fruits and those of T_{15} remained constant (Figures 2E and 2F). However, in T_{25} fruits and those of G fruits, the total sugars and reducing sugars decreased and moved further away from the Y fruit references, established as 19.49 and 13.08%, respectively (Table 1).

Vitamin C

The highest Vc (mg/100 g) at harvest were found in T fruits (49.65), followed by Y fruits (24.98), and G fruits (9.83), with significant differences among the three groups (Table 1), and no effect of temperature (Figure 3A). Except for Y_{25} , an increase in Vc was observed in the period immediately after harvest. The increase was very pronounced in T_{15} and T_{25} , where maximum values were 79.9 and 68.7 mg/100 g, respectively, while it was moderate in the rest. Thereafter, Vc (mg/100 g) decreased from 24.3 to 21.5 in G_{15} , from 79.9 to 5.3 in T_{15} , from 30.6 to 5.3 in Y_{15} , from 35.6 to 2.4 in G_{25} , from 68.7 to 3.8 in T_{25} , and from 29.9 to 2.4 in

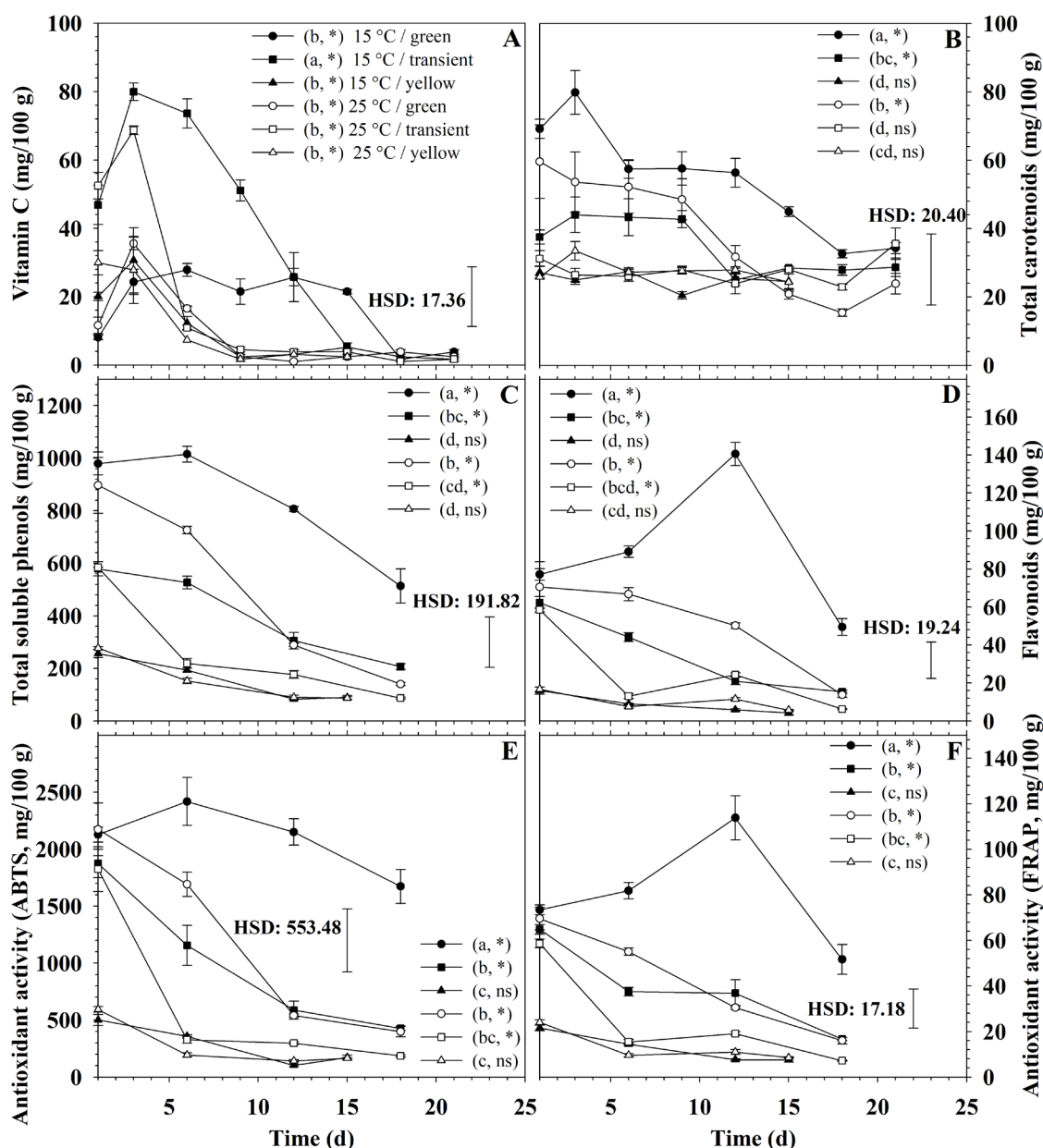


Figure 3. Contents of vitamin C, carotenoids, total soluble phenols and flavonoids, and antioxidant activity of nance fruits postharvest. Asterisk (*) indicates significant modification over time. Similar lowercase letters indicate no significant difference ($p > 0.05$) between batches. HSD: honest significant difference. Error bars indicate standard errors.

Y_{25} (Figure 3A). Similarly, Neves *et al.* (2015) reported a decrease in Vc in nance fruits from 186.5 to 4.1 mg/100 mL during storage at 15°C. Fenech *et al.* (2019) showed that Vc could increase with ripening as occurred in tomato, grape, and strawberry. However, the authors showed that Vc could also show a maximum value at an immature stage because of a biosynthetic phenomenon, as occurred in kiwi and peach fruits. Vc content is correlated with the fruit oxidative state, which is frequently detonated at the beginning of ripening. Thus, nance fruits in their transient stage could experience Vc biosynthesis as part of carbohydrate metabolism that develops mainly in association with the glycolysis pathway (Etienne *et al.*, 2013).

Carotenoids

The green or yellow hue in nance fruits is due to the presence of carotenoids and chlorophylls (Shewfelt, 2003). Although G fruits were collected at the green stage, the average initial value of hue angle was 100.9° (Table 1), which was closer to absolute yellow (90°) than to green (180°) (McGuire, 1992), thus indicating the presence of both chlorophylls and carotenoids. In fact, fruits in the green stage showed the highest values of carotenoid content at the beginning of storage (64.35 µg/100 g). This value contrasted ($p \leq 0.05$) with the content found in transient (35.35 µg/100 g) and yellow fruits (26.46 µg/100 g), which were not significantly different ($p > 0.05$) (Table 1), thus indicating a loss of these compounds with ripening, which commonly involves degradation of chlorophyll and a concomitant biosynthesis or unmasking of carotenoids (Shewfelt, 2003). Y fruits and those of T_{25} did not show significant changes ($p > 0.05$) in carotenoid content throughout storage, suggesting that no new accumulation of these compounds occurred. In contrast, G fruits and those of T_{15} experienced significant decrease ($p \leq 0.05$) in carotenoid content over time (Figure 3B), thus suggesting that changes associated with ripening occurred. All batches had the same final average value of 28.53 (± 6.56) µg/100 g, which coincided with the report of 39.4 µg/100 g by Irías-Mata *et al.* (2018). The loss of carotenoids in horticultural products during storage postharvest has been previously reported (Pandurangi and Laborde, 2004). In the present work, fruits that were not at yellow maturity experienced a decrease in the content of carotenoids; however, once they reached the mature stage, concentrations of these compounds remained approximately constant. On the other hand, the highest carotenoid contents were found at 15°C, and

the lowest at 25°C (Figure 3B). Temperature is a factor that affects carotenoid stability, and a higher thermal condition causes higher loss of these compounds during storage (Pandurangi and Laborde, 2004).

Phenolic compounds

Total soluble phenol (TSP) content was 938.05 mg/100 g in G fruits, 581.47 mg/100 g in the transient condition, and 267.14 mg/100 g in fruits at abscission maturity at the beginning of the storage (Table 1), with significant differences among maturity stages, but with no effect of temperature (Figure 3C). The TSP content of nance fruits at consumption maturity has been reported as 2,219.3 mg/100 g dry weight (dw) (Neves *et al.*, 2015), 2,900.0 mg/100 g dw (Souza *et al.*, 2008), 800 mg/100 g fresh weight (fw) (Silva *et al.*, 2007), and 334.4 mg/100 g fw (de Souza *et al.*, 2012). The TSP content can increase (Guofang *et al.*, 2019) or decrease (Hu *et al.*, 2018) with ripening, depending on fruit oxidative metabolism. In this regard, results indicated that the nance fruits in the present work experienced a decrease in TSP content with ripening. The TSP was not significantly modified after harvest in fruits at abscission maturity (Y fruits); however, it decreased in fruits harvested at green and transient maturities. In the case of G_{15} fruits, although the TSP decreased during storage, they did not reach yellow maturity, and the final TSP concentration was 513.43 (± 64.95) mg/100 g. In contrast, the rest of batches reached a final TSP content of 122.3 (± 49.7) mg/100 g, with no significant differences among them ($p > 0.05$; Figure 3C), and the value was similar to the reference value of Y fruits.

The TFC had an initial value of 73.90 mg/100 g in G fruits, 60.44 mg/100 g in transient fruits, and 16.34 mg/100 g in ripe yellow fruits, with significant differences among physiological conditions ($p \leq 0.05$) (Table 1). However, during storage, Y fruits remained without significant changes, although G_{25} and T fruits experienced a decrease in TFC, concomitant to a decrease in TSP. At the end of the study, they recorded an average value of 9.10 (± 4.91) mg/100 g, and tended toward the concentration in yellow fruits, without significant difference between batches ($p > 0.05$; Figure 3D). In G_{15} fruits, there was a temporary increment-decrement of TFC, which did not reach the reference content of fruits at abscission maturity at the end of storage, thus indicating that although there were changes associated with ripening, it was incomplete. The phenolic composition of the edible part of nance fruits was analysed by Gordon (2011),

who revealed 24 constituents including gallotannins, quinic acid gallates, proanthocyanidins, and quercetin derivatives.

Antioxidant activity

The antioxidant activity (AA) was determined with the ABTS and FRAP assays. With ABTS, initial values were 2,151.22 mg/100 g in G fruits, 1,849.10 mg/100 g in T fruits, and 547.50 mg/100 g in yellow fruits, which was significantly different from the first two ($p \leq 0.05$). With FRAP, initial values were 71.47, 61.72, and 22.78 mg/100 g, respectively, with significant difference among the three physiological states (Table 1). Differences between methods occurred because the ABTS assay is based on electron transfer evaluation, while the FRAP assay focuses on hydrogen atom transfer (Apak *et al.*, 2016).

Vitamin C, carotenoids, and phenolic compounds are important factors in AA, and data corresponding to the immediate stage after harvest indicated that these compounds, as well as antioxidant capacity, decreased with ripening (Table 1). Bioactive compounds in nance fruits can have beneficial effects on health (Herrera-Ruiz *et al.*, 2011); however, the leaves and barks have been reported to have higher values than fruits. Silva *et al.* (2007) determined values of 347.1, 261.3, and 4.2 in leaves, barks, and fruits with the TEAC assay, and 778.8, 590.8, and 11.8 μmol equivalent of Trolox (TE) per gram fw, respectively, with the ORAC assay. Also, Souza *et al.* (2008) reported 736.0, 1145.0, and 26.5 μmol TE/g dw in leaves, barks, and fruits, respectively, thus confirming that AA of fruits is lower than that of other plant parts. Based on Figures 3E and 3F, when postharvest handling occurred at 15°C, AA was higher than at 25°C, at a ratio of 11.3/7.1 with ABTS, and 4.4/2.7 with FRAP. AA of Y fruits was not significantly modified during storage ($p > 0.05$). However, like TSP, the AA of G_{25} fruits and that of T fruits decreased and, at the end of the study, values like those of fruits at abscission maturity remained, with an average value of 295.72 (± 33.34) mg/100 g based on ABTS assay, and 11.20 (± 4.58) mg/100 g based on FRAP assay. AA decreased in G_{15} fruits, and final values contrasted with the rest, thus suggesting that a trend towards ripening occurred, but it was not fully completed (Figure 3E and 3F).

Feasibility of early harvest

Maturity at harvest determines the behaviour throughout storage. Y fruits started storage at abscission maturity, corresponding to senescence,

and behaviour during storage was characterised by excessive softening, decrease in hue angle to brown tones, and loss of lightness and chroma. In addition, because ripening had already elapsed, compositional variables remained without significant modification regardless of the temperature condition. Acidity was an exception, and some batches experienced an increase as storage progressed, possibly caused by microbial growth. The quality attributes of fruits at abscission maturity (Y) were the reference to evaluate postharvest behaviour of fruits harvested at green (G) and transient (T) stages. Although G fruits reached the reference firmness, in general, they did not develop adequate colour attributes, and did not reach the reference contents of TSS and sugars. This indicated that adequate sensory attributes were not developed, and classification of the fruit as non-climacteric was strengthened. Although these fruits retained high values of compounds associated with antioxidant activity, particularly at 15°C, harvest at this maturity stage did not favour adequate ripening. Anami *et al.* (2020) showed that the handling of strawberry, a non-climacteric fruit, harvested at 1/3 ripe condition, allowed the antioxidant potential to have higher values, but colour attributes did not reach typical consumption values, as did fruits that were harvested at 3/4 ripeness. In the present work, fruits harvested at transient maturity experienced softening, thus giving them a texture similar to that of yellow fruits, and the hue angle and chroma tended toward the reference values, although lightness decreased. TSS content remained without significant changes, although sugar content did not increase up to the reference values. Handling at 15°C allowed these compounds to be preserved, in contrast to handling at 25°C, where there was a notable decrease in them. In addition, contents of soluble phenols, flavonoids, carotenoids, and antioxidant activity decreased in a way similar to fruits at abscission maturity. On the other hand, the use of refrigeration at 15°C caused changes to occur at a slower rate, which suggested possible shelf life prolongation. Rivas-Castro *et al.* (2019) argued that nance fruits can be harvested at a stage prior to abscission maturity. Results obtained in the present work showed that fruits harvested at the transient maturity stage could undergo some changes that lead to ripening after they were separated from the tree, and could be handled for more than 15 d under the refrigerated conditions applied. However, it is necessary to determine the stage of maturity at which fruits are harvested that favours complete ripening to achieve the quality attributes of fruits at consumption maturity.

Conclusion

Nance fruits showed non-climacteric behaviour. However, if they were harvested at a physiological state prior to consumption maturity, they might undergo changes associated with ripening. Harvesting nance fruits in the transient stage, between physiological and consumption maturities, and handling under refrigeration at 15°C could extend shelf life for more than 15 d, with adequate physical and compositional attributes, including high concentration of bioactive compounds and antioxidant activity. However, actions should be taken to control fungal development and to determine an adequate harvest index, which can be based on total soluble solids.

Acknowledgement

Authors wish to acknowledge the financial support received from the scholarship program of Consejo Nacional de Ciencia y Tecnología (CONACyT) of Mexico.

References

- Anami, J. M., Steffens, C. A., Moreira, M. A., Fernandes, R. C., Mosquera, D. J. C. and Do Amarante, C. V. T. 2020. Active modified atmosphere storage to preserve the quality of 'San Andreas' strawberries harvest at two ripening stages. *Revista Brasileira de Fruticultura* 42(1): 1-10.
- Apak, R., Özyürek, M., Güçlü, K. and Çapanoğlu, E. 2016. Antioxidant activity/capacity measurement. 1. Classification, physicochemical principles, mechanisms, and electron transfer (ET)-based assays. *Journal of Agricultural and Food Chemistry* 64(5): 997-1027.
- Association of Official Analytical Chemist (AOAC). 1990. Official methods of analysis of AOAC International. 15th ed. United States: AOAC.
- Benzie, I. F. F. and Strain, J. J. 1996. The ferric reducing ability of plasma (FRAP) as a measure of "antioxidant power": the FRAP assay. *Analytical Biochemistry* 239(1): 70-76.
- Cao, W., Zhang, J. J., Liu, C. Y., Bai, W. S. and Cheng, N. 2020. A modified Folin-Ciocalteu method for the microdetermination of total phenolic content in honey. *International Food Research Journal* 27(3): 576-584.
- Chang, C. C., Yang, M. H., Wen, H. M. and Chern, J. C. 2002. Estimation of total flavonoid content in propolis by two complementary colometric methods. *Journal of Food and Drug Analysis* 10(3): 178-182.
- Cherian, S., Figueroa, C. R. and Nair, H. 2014. "Movers and shakers" in the regulation of fruit ripening: a cross-dissection of climacteric versus non-climacteric fruit. *Journal of Experimental Botany* 65(17): 4705-4722.
- da Silva, M. C., Neves, L. C., Bastos, V. J., Lima, C. G. B., De Araújo, K. G. M. and Roberto, S. R. 2016. Harvesting period of Murici (*Byrsonima crassifolia* Kunth) fruit in relation to physical and chemical parameters evaluated during fruit development. *Scientia Horticulturae* 200(1): 66-72.
- de Souza, V. R., Aparecida, P., Pereira, P., Queiroz, F. and Borges, S. V. 2012. Determination of bioactive compounds, antioxidant activity and chemical composition of Cerrado Brazilian fruits. *Food Chemistry* 134(1): 381-386.
- Duarte, O. 2011. Nance (*Byrsonima crassifolia* (L.) Kunth). In Yahia, E. M. (ed). *Postharvest Biology and Technology of Tropical and Subtropical Fruits, Volume 4 - Mangosteen to White Sapote*, p. 44-50. United Kingdom: Woodhead Publishing Limited.
- Duarte, O. and Paull, R. 2015. *Exotic fruits and nuts of the new world*. Boston: CABI.
- Dudonné, S., Vitrac, X., Coutière, P., Woillez, M. and Mérillon, J. M. 2009. Comparative study of antioxidant properties and total phenolic content of 30 plant extracts of industrial interest using DPPH, ABTS, FRAP, SOD, and ORAC assays. *Journal of Agricultural and Food Chemistry* 57: 1768-1774.
- Etienne, A., Génard, M., Lobit, P., Mbéguié-A-Mbéguié, D. and Bugaud, C. 2013. What controls fleshy fruit acidity? A review of malate and citrate accumulation in fruit cells. *Journal of Experimental Botany* 64(6): 1451-1469.
- Fenech, M., Amaya, I., Valpuesta, V. and Botella M. A. 2019. Vitamin C content in fruits: biosynthesis and regulation. *Frontiers in Plant Science* 9(1): 1-21.
- Gordon, A. 2011. *Bioactive compounds in underutilized tropical fruits from Latin America*. Germany: University of Bonn, PhD thesis.
- Guofang, X., Xiaoyan, X., Xiaoli, Z., Yongling, L. and Zhibing, Z. 2019. Changes in phenolic profiles and antioxidant activity in rabbiteye blueberries during ripening. *International Journal of Food Properties* 22(1): 320-329.
- Herrera-Ruiz, M., Zamilpa, A., González-Cortazar, M., Reyes-Chilpa, R., León, E., García, M. P., ... and Huerta-Reyes, M. 2011. Antidepressant

- effect and pharmacological evaluation of standardized extract of flavonoids from *Byrsonima crassifolia*. *Phytomedicine* 18(14): 1255-1261.
- Hu, K., Dars, A. G., Liu, Q., Xie, B. and Sun, Z. 2018. Phytochemical profiling of the ripening of Chinese mango (*Mangifera indica* L.) cultivars by real-time monitoring using UPLC-ESI-QTOF-MS and its potential benefits as prebiotic ingredients. *Food Chemistry* 256(1): 171-180.
- Huang, Z., Hu, H., Shen, F., Wu, B., Wang, X., Zhang, B., ... and Zhang, X. 2018. Relatively high acidity is an important breeding objective for fresh juice-specific apple cultivars. *Scientia Horticulturae* 233: 29-37.
- Irías-Mata, A., Jiménez, V. M., Björn, C., Schweigert, R. M., Carle, R. and Esquivel, P. 2018. Carotenoids and xanthophyll esters of yellow and red nance fruits (*Byrsonima crassifolia* (L.) Kunth) from Costa Rica. *Food Research International* 111: 708-714.
- Kader, A. A. 2002. Postharvest biology and technology: an overview. In Kader A. A. (ed). *Postharvest Technology of Horticultural Crops*, p. 39-47. Oakland: University of California.
- Mahajan, P. V., Luca, A. and Edelenbos, M. 2014. Impact of mixtures of different fresh-cut fruits on respiration and ethylene production rates. *Journal of Food Science* 79(7): E1366-E1371.
- Mariutti, L. R. B., Rodrigues, E., Chisté, R. C., Fernandes, E. and Mercadante, A. Z. 2014. The Amazonian fruit *Byrsonima crassifolia* effectively scavenges reactive oxygen and nitrogen species and protects human erythrocytes against oxidative damage. *Food Research International* 64(1): 618-625.
- McGuire, R. G. 1992. Reporting of objective color measurements. *HortScience* 27(12): 1254-1255.
- Medina-Torres, R., Salazar-García, S. and Gómez-Aguilar, J. R. 2004. Fruit quality indices in eight nance [*Byrsonima crassifolia* (L.) H.B.K.] selections. *HortScience* 39(5): 1070-1073.
- Medina-Torres, R., Salazar-García, S., Valdivia-Bernal, R. and Martínez-Moreno, E. 2012. Flowering phenology and reproductive cycles of nance [*Byrsonima crassifolia* (L.) HBK] in Nayarit. *Universidad y Ciencia* 28(3): 259-269.
- Miller, G. L. 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Analytical Chemistry* 31(3): 426-428.
- Miranda-Molina, F. D., Valle-Guadarrama, S., Guerra-Ramírez, D., Arévalo-Galarza, M. D. L., Pérez-Grajales, M. and Artés-Hernández, F. 2019. Quality attributes and antioxidant properties of Serrano chili peppers (*Capsicum annuum* L.) affected by thermal conditions postharvest. *International Food Research Journal* 26(6): 1889-1898.
- Moo-Huchin, V. M., Estrada-Mota, I., Estrada-León, R., Cuevas-Glory, L., Ortiz-Vázquez, E., Vargas, V. M. L., ... and Sauri-Duch, E. 2014. Determination of some physicochemical characteristics, bioactive compounds and antioxidant activity of tropical fruits from Yucatan, Mexico. *Food Chemistry* 152(1): 508-515.
- Neves, L. C., Tosin, J. M., Benedette, R. M. and Cisneros-Zevallos, L. 2015. Post-harvest nutraceutical behaviour during ripening and senescence of 8 highly perishable fruit species from the Northern Brazilian Amazon region. *Food Chemistry* 174(1): 188-196.
- Nielsen, S. S. 2010. Vitamin C determination by indophenol method. In Nielsen, S. S. (ed). *Food Analysis Laboratory Manual*, p. 55-60. United States: Springer.
- Nunes, M. C. N. and Emond, J. P. 2003. Storage temperature. In Bartz, J. A. and Brecht, J. K. (eds). *Postharvest Physiology and Pathology of Vegetables*, p. 209-228. New York: Marcel Dekker.
- Pandurangi, S. and Laborde, L. F. 2004. Retention of folate, carotenoids, and other quality characteristics in commercially packaged fresh spinach. *Journal of Food Science* 69(9): C702-C707.
- Pérez, G. R. M. and Muñiz, R. A. 2016. Hypoglycemic effects of sesquiterpene lactones from *Byrsonima crassifolia*. *Food Science and Biotechnology* 25(4): 1135-1145.
- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M. and Rice-Evans, C. 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine* 26(9-10): 1231-1237.
- Rivas-Castro, S. F., Martínez-Moreno, E., Alia-Tejaca, I. and Pérez-López, A. 2019. Physical and physiological changes in phenotypes of nance [*Byrsonima crassifolia* (L.) H.B.K.] with different harvest maturity. *Scientia Horticulturae* 256(1): article ID 108620.
- Safařík, I. V. O. and Šantrůčková, H. 1992. Direct determination of total soil carbohydrate content. *Plant and Soil* 143: 109-114.
- Séfora, B. S. M., Marques, L. J. J. and De Souza, B. D. 2019. Optimization of the extraction of polyphenols and antioxidant capacity from *Byrsonima crassifolia* (L.) Kunth fruit by

- response surface methodology. In Soto-Hernández, R. M., García-Mateos, R. and Palma-Tenango, M. (eds). *Plant Physiological Aspects of Phenolic Compounds*, p. 1-12. London: IntechOpen.
- Shewfelt, R. L. 2003. Color. In Bartz, J. A. and Brecht, J. K. (eds). *Postharvest Physiology and Pathology of Vegetables*, p. 287-296. New York: Marcel Dekker.
- Silva, E. M., Souza, J. N. S., Rogez, H., Rees, J. F. and Larondelle, Y. 2007. Antioxidant activities and polyphenolic contents of fifteen selected plant species from the Amazonian region. *Food Chemistry* 101(3): 1012-1018.
- Singleton, V. L. and Rossi, J. J. A. 1965. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *American Journal of Enology and Viticulture* 16: 144-158.
- Souza, J. N. S., Silva, E. M., Loir, A., Rees, J. F., Rogez, H. and Larondelle, Y. 2008. Antioxidant capacity of four polyphenol-rich Amazonian plant extracts: a correlation study using chemical and biological in vitro assays. *Food Chemistry* 106(1): 331-339.
- Speek, A. J., Speek-Saichua, S. and Schreurs, W. H. P. 1988. Total carotenoid and β -carotene contents of Thai vegetables and the effect of processing. *Food Chemistry* 27(4): 245-257.
- Valle-Guadarrama, S., Morales-Cabrera, M., Peña-Valdivia, C. B., Mora-Rodríguez, B., I. Alia-Tejacal, I., Corrales-García, J. and Gómez-Cruz, A. 2013. Oxidative/fermentative behavior in the flesh of "Hass" avocado fruits under natural and controlled atmosphere conditions. *Food and Bioprocess Technology* 6: 272-282.
- Wang, D., Yeats, T. H., Uluisik, S., Rose, J. K. C. and Seymour G. B. 2018. Fruit softening: revisiting the role of pectin. *Trends in Plant Science* 23(4): 302-310.