Effect of soaking in noni (*Morinda citrifolia*) juice on the microbiological and color behavior of Haden minimally processed mango

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Abstract The purpose of this study was to evaluate the effect of soaking in noni juice on the microbiological and color behavior of minimally processed mango. Two batches of Haden mango cubes were treated by immersion in noni juice for 2.5 or 5.0 min. Each batch was packed in polypropylene boxes and stored at 6 °C for up to 15 days; in addition, a control group of mango cubes was prepared by immersion in sterile water for the same duration. According to the results, the soaking of mango cubes in noni juice had an antimicrobial effect on mesophilic aerobic bacteria, molds and yeasts during storage at 6 °C for 15 days, without significantly ($P < 0.05$) affecting the CIE L^* , a^* , b^* , chroma and hue angle values, in comparison with the control after 12 days of storage. The noni juice soaking treatment was demonstrated to be a potentially valuable technology for decontamination of fresh-cut fruit surfaces.

Keywords Noni juice · Antimicrobial activity · Fresh-cut fruit · Haden mango · Color · *Morinda citrifolia*

Introduction

In recent times, the consumption of fruits and vegetables has gradually increased year by year. People are now more aware of the importance of a healthy diet and consumers require quality and convenience in the products they buy. Thus, minimally processed fruits and vegetables can be considered an alternative to fast food and other ready-to-eat products. Apart from satisfying the demand for healthy and convenient food, they may permit better use of raw materials, and offer the possibility of increasing the product value (González-Aguilar et al. 2005; Luo 2007; Sabir et al. 2011).

Preparation of fresh-cut fruits and vegetables generally consists of washing, cutting, treatment with sanitizing agents, packaging of products and storage under refrigerated conditions (McKellar et al. 2004). Cutting or slicing operations induce a reduction in shelf life by modifying the metabolic processes of vegetal tissue and increasing its susceptibility to spoilage (Raybaudi-Massilia et al. 2009) and browning reactions as a result of the compartmentalization loss of phenolic compounds (mainly in the vacuole) and polyphenol oxidase (in the cytoplasm) (Sapers and Miller 1998; Soliva-Fortuny and Martín-Belloso 2003; Rico et al. 2006; Del Nobile et al. 2009).

In the case of fresh-cut mangoes, browning, softening, surface dehydration, water loss, translucency, off-flavour and off-odour development, as well as microbial spoilage are some of the most frequent causes of quality loss (Siddiq et al. 2013; Sellamuthu et al. 2013).

Chemical synthetic additives can reduce the decay rate, but consumers are concerned about chemical residues in the product that could affect their health and cause environmental

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pollution (Wang 2006; Ayala-Zavala et al. 2008a). Therefore, alternative methods for controlling fresh-cut fruit decay are required.

One of the major emerging technologies for reducing quality loss and increasing the safety of fresh-cut fruits and vegetables is the application of natural additives (Ponce et al. 2004; Ayala-Zavala et al. 2008b). Plants and plant products represent a source of natural antioxidants and antimicrobials to improve the shelf life and safety of foods including fresh-cut fruit (Cuppett et al. 1997; Donovan et al. 1998; Chouliara et al. 2007; Solomakos et al. 2008; Espina et al. 2011; Dahech et al. 2013; Senthilkumar and Venkatesalu 2013).

Morinda citrifolia (noni) has been used in folk remedies by Polynesians for over 2,000 years, and is reported to have a broad range of therapeutic effects, including antibacterial, antiviral, antifungal, antitumour, antihelmintic, analgesic, hypotensive, anti-inflammatory, and immune-enhancing effects (Wang et al. 2002; Pawlus and Kinghorn 2007). About 200 phytochemicals have been identified from the noni plant, including anthraquinones, flavonoids, polysaccharides, glycosides, iridoids, lignans, organic acids, triterpenoids, and organic acids (Wang and Su 2001; Chan-Blanco et al. 2006; Pawlus and Kinghorn 2007; Deng et al. 2010). A total of 51 volatile compounds have also been found in the ripe noni fruit including organic acids (Farine et al. 1996; Sang et al. 2001).

It has been reported that noni extracts inhibit the growth of certain bacteria, such as *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Proteus morganii*, *Bacillus subtilis*, *Escherichia coli*, *Helicobacter pylori*, *Streptococcus pyogenes*, *Salmonella* spp. and *Shigella* spp. (Dittmar 1993; Locher et al. 1995; Leach et al. 1988). This antimicrobial effect may be due to the presence of phenolic compounds such as acubin, L-asperuloside, alizarin, scopoletin and other anthraquinones (Atkinson 1956). It has also been found that ethanol and hexane extracts of noni have an antitubercular effect since they inhibit the growth of *Mycobacterium tuberculosis* by 89–95 % (Saludes et al. 2002). The antimicrobial properties of noni extracts have been used for the treatment of some skin infections, colds, fevers and other bacterial-related health problems (Atkinson 1956), but not for applications in the preservation of foods.

On the other hand, modern scientific research has shown that noni fruits possess antioxidant effects that prevent and cure several diseases (Deng et al. 2010; Kumoro et al. 2011; Fabra et al. 2011). According to Tapp et al. (2012), the natural antioxidant properties of noni have the potential to prevent lipid oxidation and improve color, while other studies suggest that extracts of noni might be useful as a natural and multifunctional dietary food additive or supplement because of its high antioxidant activity (Zin et al. 2007; Krishnaiah et al. 2012).

Therefore, this study focused on determining the effects of soaking in naturally fermented noni juice on the microbiological characteristics and color behavior of minimally processed mango during storage at 6 °C.

Materials and methods

Noni fruit and juice preparation

Noni fruit was obtained from a plantation of noni trees located at El Llano, municipality of San Blas, Nayarit, México. The collected noni fruit was transported to the laboratory of the Centro de Tecnología de Alimentos of the Universidad Autónoma de Nayarit, based in Tepic, Nayarit, México. Then, the noni fruit was sorted and washed with tap water. The washed noni fruit was placed in sealed plastic jars (10 L) to obtain noni juice via the traditional method by natural fermentation at an ambient temperature of 25 °C for 60 days (Nelson and Elevitch 2006). Noni juice that dripped from the fruits was collected and centrifuged at 3,050×g in a Solbat model 2,774 centrifuge (Solbat Aparatos Científicos, Puebla, México) for 15 min. The supernatant was decanted and bottled in dark jars as fresh noni juice, which were kept at room temperature (25 °C) in darkness to be used 10 days later in this study.

Mango and cube preparation

Haden mangoes were obtained from a farmer who lives in the Navarrete municipality of San Blas, Nayarit, México, and transported to the Centro de Tecnología de Alimentos of the Universidad Autónoma de Nayarit. Mature green mangoes were picked in anticipation of the processing date and were allowed to ripen for 4 days at room temperature (25 °C) and natural light. The mangoes were washed with tap water and peeled with a knife; the flesh was removed from the seed, and was then cut into 2-cm³ cubes.

Soaking mango cubes in noni juice

Immersion treatments of 2.5 and 5.0 min were carried out to evaluate the effect of soaking in noni juice at room temperature (25 °C) on the microbiological characteristics of minimally processed mango. The mango cubes (about 250 g) from each treatment were drained and placed in polypropylene boxes. In addition, mango cube control treatments were prepared by immersion in sterile water for the same durations as the treatments in noni juice. After the experimental treatments, all mango cubes were stored in a refrigerator at 6 °C in darkness for 15 days.

Microbial counts

Serial solutions were made and poured onto duplicate Standard Method Agar (Bioxon, Cuatitlán Izcalli, México) plates, which were incubated at 37 °C for 48 h for enumeration of total bacteria, and onto duplicate Potato Dextrose Agar plates (Bioxon, Cuatitlán Izcalli, México) with 100 mg/L

chloramphenicol (Sigma-Aldrich, St. Louis, MO, USA), which were incubated at 26 °C for 5–7 days for evaluation of yeast and molds. Total microbial counts were determined every 3 days and expressed as log colony forming units (log cfu/g).

Physicochemical analysis

Titrate acidity, pH, total soluble solids (°Brix) and total and reducing sugars were determined according to the official methods of the Association of Official Analytical Chemists (AOAC 1984). Measurements of pH were done with a Denver Instrument model 250 pH meter (Denver Instrument Company, Denver, CO, USA) and °Brix on an Auto Abbe model 10500 refractometer (Reichert-Jung, Leica Inc., Buffalo, NY, USA). Water activity measurements were performed using an Aqua Lab CX2 water activity meter (Decagon Devices Inc, Pullman, WA, USA). Color characteristics were measured using a ColorQuest colorimeter (HunterLab; Hunter Associates Laboratories Inc., Reston, VA, USA) with the specular component included, Illuminant C, and 10° observer angle parameters set for transmittance on noni juice and reflectance on mango samples. L^* (lightness), a^* (redness and greenness), and b^* (yellowness and blueness) were measured based on CIE tri-stimulus color space. The colorimeter was always calibrated using a white tile before analysis. Chroma (C^*) was calculated as: $C^* = [(a^*)^2 + (b^*)^2]^{1/2}$ for the intensity or color saturation. Hue angle (h°) was calculated as $\tan^{-1} b^*/a^*$ and expressed in degrees as the attribute according to which colors have been traditionally defined as reddish, greenish, etc.

Statistical analyses

Each microbiological value was the mean of the counts carried out on three samples of each experiment repeated three times from different groups after the same storage period. Differences among the microbial counts were analyzed by one-way analysis of variance (ANOVA) using the statistical package Statgraphics Plus Version 4 (Manugistics Inc., Rockville, MD, USA). The mean values were compared using LSD multiple range tests. Prior to ANOVA, all cfu/g data were transformed to $\log_{10}$ cfu/g. Differences among treatments were based on a significance level of $P < 0.05$.

Results and discussion

Table 1 shows the physicochemical and microbiological characteristics of the noni juice obtained by natural fermentation according to the traditional method. The values for °Brix, titratable acidity, reducing sugars and pH of the noni juice

Table 1 Some physicochemical and microbial characteristics of noni juice (*Morinda citrifolia*)

Characteristic	^a Value
Total soluble solids (°Brix)	5.6±0.1
Titrate acidity (g/L)	3.04±0.1
Reducing sugars (g/L)	4.38±0.1
Total sugars (g/L)	5.53±0.1
pH	3.16±0.05
Color	
L^* (lightness)	18.39±0.09
a^* (redness-greenness)	2.15±0.02
b^* (yellowness-blueness)	8.88±0.17
C^* (chroma)	8.55±0.32
h° (hue angle)	14.64±0.67
Water activity	0.991±0.001
Mesophilic aerobic bacteria (log cfu/mL)	<1.0
Molds (log cfu/mL)	<1.0
Yeasts (log cfu/mL)	<1.0

^a Values are given as mean ± standard deviation of one sample by triplicate

from this study were lower than those reported by Satwadhara et al. (2011). With respect to the first attribute of color, lightness (L^*), which is associated with the luminous intensity and describes the light-reflecting or -transmitting capacity of an object, the noni juice showed a low value typical of the juices from this fruit, while the values of a^* and b^* , which are associated with red and yellow colors, respectively, were positive (Zulueta et al. 2007). Microbial counts of mesophilic aerobic bacteria, molds and yeasts were not detected in the noni juice.

Microbiological characteristics

Table 2 shows the effect of soaking in noni juice on the microbiological characteristics of mango cubes stored at 6 °C over 15 days. According to the results, the microbial counts of mesophilic aerobic bacteria, molds and yeasts on mango cubes showed a significant ($P < 0.05$) reduction after soaking in noni juice, in contrast with the control groups, which showed an increase, especially for mesophilic aerobic bacteria and yeasts. However, aerobic mesophilic bacterial count decreased at 3rd day in case of control mango cubes and thereafter increased again, which is characteristic of the lag phase because cells undergo intracellular changes in an effort to adjust to a new environment (Yates and Smotzer 2007), as in this case to the storage temperature of 6 °C. On the other hand, the mango cubes soaked in noni juice for 5.0 min showed significantly ($P < 0.05$) lower microbial counts during storage at 6 °C than mango cubes soaked in noni juice for 2.5 min.

Table 2 Effect of soaking in noni juice on microbial behavior (log cfu/g) of mango cubes stored at 6 °C

Microbial group	Soaking time (min)	Storage time (days) ^a					
		0	3	6	9	12	15
Aerobic mesophilic bacteria	0 (control)	3.83±0.02aA	2.48±0.05bA	3.11±0.00cA	3.93±0.02dA	3.97±0.01eA	4.47±0.06fA
	2.5	2.93±0.04aB	2.60±0.08bB	1.02±0.03cB	1.02±0.03cB	1.02±0.03cB	1.02±0.03cB
	5.0	1.81±0.05aC	1.76±0.03bC	1.02±0.03cB	0.98±0.03dC	0.93±0.04dC	0.87±0.04eC
Molds	0 (control)	1.31±0.01aA	1.69±0.12bA	1.02±0.03cA	1.02±0.03cA	1.02±0.03cA	1.02±0.03cA
	2.5	1.02±0.01aB	0.98±0.02bB	0.93±0.02cB	0.81±0.02 dB	0.75±0.03eB	0.65±0.04fB
	5.0	0.98±0.03aC	0.93±0.04bC	0.87±0.04cC	0.81±0.05 dB	0.65±0.06eC	0.54±0.07fC
Yeasts	0 (control)	2.40±0.27aA	2.88±0.06bA	4.08±0.01cA	4.08±0.05cA	4.56±0.04dA	4.63±0.05dA
	2.5	1.02±0.03aB	0.98±0.03bB	0.93±0.04cB	0.93±0.04cB	0.87±0.04 dB	0.81±0.05 dB
	5.0	0.98±0.03aC	0.93±0.04bC	0.87±0.04cC	0.81±0.05dC	0.74±0.06eC	0.65±0.07fC

^a Values are averages of log cfu/g ± standard deviation of three samples measured in each experiment repeated three times. Mean values within the same line followed by the same lowercase letter are not significantly different ($P<0.05$) based on a LSD multiple rank test. Mean values within the same column for each microbial group followed by the same uppercase letter are not significantly different ($P<0.05$) based on LSD's multiple rank test

The effects of some treatments on the microbiological behavior of fresh-cut mangoes have been reported. Chiumarelli et al. (2010) studied the effect of citric acid dipping and cassava starch coating, with and without glycerol, on quality preservation of fresh-cut mangoes during storage at 5 °C for 15 days, finding that such treatments resulted in yeast and mold counts around 4–5 log cfu/g. Moreover, a controlled atmosphere of 2 % O₂ and 10 % CO₂ maintained the initial total bacterial and yeast/mold counts of Tommy Atkins and Kent mango cubes stored at 5 °C for 8 days; however, when the mango cubes were treated

with the same controlled atmosphere conditions but stored at 10 °C, the initial total bacterial and yeast/mold counts increased by about 1 log cfu/g (Rattanapanone et al. 2001). In contrast with the results described above, the noni juice soaking treatments for 2.5 and 5.0 min applied to mango cubes in this study demonstrated antimicrobial activity during storage at 6 °C for 15 days through a reduction in microbial counts of about 3.6 log cfu/g for total mesophilic aerobic bacteria, 4 log cfu/g for yeasts and 0.5 log cfu/g for moulds, may be due to the presence of natural antioxidants such as phenolic compounds, mainly of

Table 3 Effect of soaking in noni juice on color behavior of mango cubes stored at 6 °C

Color parameter	Time soaking (min)	Storage time (days) ^a					
		0	3	6	9	12	15
<i>L</i> *	0 (control)	63.62±7.96aA	61.68±2.94aA	61.70±4.28aA	59.45±3.79aA	61.43±5.39aA	59.94±7.55aA
	2.5	59.21±3.85abA	53.97±6.66bcB	60.04±4.87abA	59.49±5.71abA	62.44±6.59aA	47.86±6.35cB
	5.0	62.43±4.82abA	57.11±4.27bcAB	53.19±2.39cB	56.22±5.05cA	63.37±7.37aA	55.64±4.99cAB
<i>a</i> *	0 (control)	−0.61±2.98aA	1.03±1.78abA	0.10±1.63abA	−0.21±2.09abA	1.80±1.43bA	1.06±1.76abA
	2.5	1.01±2.22aA	1.24±1.03aA	0.41±1.33aA	0.06±1.69aA	0.07±2.13aA	−0.76±2.75aA
	5.0	1.73±2.36aA	0.64±1.03abA	2.31±1.44bB	0.22±1.53abA	0.58±3.75abA	−1.06±3.22abA
<i>b</i> *	0 (control)	72.63±8.66aA	71.55±5.90abA	70.98±3.98abA	67.24±6.28abA	67.93±5.97abA	64.01±7.76bA
	2.5	67.79±5.00aA	59.88±9.97aB	67.00±6.23aA	67.13±6.61aA	67.56±4.41aA	50.94±8.19bB
	5.0	65.96±5.10abA	64.21±5.55abcAB	58.78±2.02cB	62.41±5.12bcA	69.36±6.54aA	59.13±6.14cAB
Chroma	0 (control)	72.68±8.68aA	71.58±5.88abA	70.99±3.99abA	67.27±5.94abA	67.97±5.94abA	64.04±7.77bA
	2.5	67.83±4.95aA	59.90±9.07aB	67.01±6.24aA	67.14±6.62aA	67.59±4.42aA	51.01±8.13bB
	5.0	69.68±4.43aA	64.22±5.54abcAB	58.84±2.03bB	62.42±5.12bA	69.45±6.48abA	59.22±6.06bAB
Hue angle	0 (control)	90.35±2.28aA	89.10±1.47abA	89.91±1.28abA	90.19±1.82abA	88.41±1.28bA	89.07±1.49abA
	2.5	89.06±2.04aA	89.62±1.62aA	89.63±1.08aA	90.00±1.36aA	90.00±1.76aA	91.15±3.39aA
	5.0	88.49±2.06abA	89.42±0.93abA	87.75±1.37aB	89.77±1.34abA	89.35±3.24abA	91.13±3.36bA

^a Values are averages of color parameter ± standard deviation of three samples measured in each experiment repeated three times. Mean values within the same line followed by the same lowercase letter are not significantly different ($P<0.05$) based on an LSD multiple rank test. Mean values within the same column followed by the same uppercase letter for each color parameter are not significantly different ($P<0.05$) based on an LSD multiple rank test

acubin, L-asperuloside, alizarin, scopoletin and other anthraquinones (Chan-Blanco et al. 2006; Yang et al. 2010).

Color behavior

Color parameters are also important quality indexes for fresh-cut fruits. Table 3 shows the color changes (L^* , a^* , b^* , h° , and C^*) of mango cubes soaked in noni juice for 2.5 and 5.0 min and stored at 6 °C over 15 days. Until the 12th day of storage, the L^* values of mango cubes soaked in noni juice for 2.5 and 5.0 min did not show significant ($P<0.05$) differences in comparison with the initial L^* values; however, a variable behavior was observed in the L^* values for both treatments during a storage period of 3–9 days. On the 15th day of storage, reductions of 19.2 % and 10.9 % on the L^* values were observed for mango cubes soaked in noni juice for 2.5 and 5.0 min, respectively.

A 15.9 % reduction in the L^* value was observed for Ataulfo mango cubes stored at 5 °C for 15 days after dipping treatment in an aqueous solution of 1 % ascorbic acid + citric acid + calcium chloride for 3 min, which was subsequently drained (Robles-Sánchez et al. 2009). Another study with Kent fresh-cut mango slices showed reductions of 9.2 %, 12.3 %, 18.5 % and 20.0 % in the L^* initial value after 13 days of storage (5 °C) by (i) soaking in an anti-browning mix (calcium ascorbate, citric acid, and N-acetyl-L-cysteine), soaking in anti-browning mix plus coating in (ii) carboxymethylcellulose, maltodextrin or (iii) carrageenan, and (iv) chitosan coating, respectively (Plotto et al. 2010). The reductions in L^* values for mango cubes during storage after soaking in noni juice in this study are in the same range as the reductions for the different treatments applied to fresh-cut mango described above.

L^* and b^* values are indicators of lightness and yellow color in fruits, respectively. In particular, the L^* value has been used for fresh-cut mango as a good indicator of surface browning in response to peeling or slicing, which facilitates the contact of oxygen and substrates with browning enzymes (Soliva-Fortuny and Martín-Belloso 2003). For fresh-cut mangoes, a lower L^* value can be an indicator of flesh browning, while increases in the a^* and b^* values indicate flesh becoming more red/orange and yellow, respectively (González-Aguilar et al. 2000). In this experiment, the a^* values of the control and the mango cubes soaked in noni juice (2.5 and 5.0 min) were not significantly ($P<0.05$) different for most of the storage periods, and no significant ($P<0.05$) differences were found in any storage period, except for the 6th day. For the b^* values, the control and the mango cubes soaked in noni juice for 2.5 min showed the same behavior up to the 12th day of storage without significant ($P<0.05$) differences with respect to the initial values. On the other hand, the control and the mango cubes soaked in noni juice for 5.0 min did not show significant

($P<0.05$) differences in any storage period during the experiment, except for the 6th day (Table 3). In contrast, the dipping treatment with ascorbic acid, citric acid and calcium chloride of Ataulfo fresh-cut mango provoked a change in the b^* value of 20 units during storage for 15 days at 5 °C (Robles-Sánchez et al. 2009).

According to Pomeranz and Meloan (1994), C^* is a measure of saturation, the purity or intensity of color. Thus, changes in C^* values are related to the ripening process and processing effects. In the present study, the control and the mango cubes soaked in noni juice for 2.5 min did not show significant ($P<0.05$) differences for C^* between storage periods except for those stored for 12 days (Table 3). Fresh-cut Tommy Atkins mango treated with a combination of citric acid dipping and coating with cassava starch and glycerol, stored at 5 °C for 9 days, showed a reduction range of 10–20 units of C^* (Chiumarelli et al. 2010).

Plotto et al. (2006) used h° as an indicator of flesh color shift from light yellow to orange/red as values decrease in stored cut mangoes. In this study, the h° of the control and the mango cubes soaked in noni juice (2.5 and 5.0 min) did not show ($P<0.05$) significant differences at the end of the experiment (Table 3).

Conclusions

According to the results obtained in this study, soaking in noni juice had a beneficial effect on the microbiological and color properties of mango cubes during storage. The 5.0 min noni juice soaking treatment applied to mango cubes stored for 15 days at 6 °C gave the best results in terms of antimicrobial effect and maintenance of color.

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