## Minimally processed yellow melon enriched with probiotic bacteria

## Melão minimamente processado enriquecido com bactéria probiótica

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## Abstract

The demand for healthy diets with fresh foods, especially minimally processed fruits and vegetables, resulted in a variety of products available to consumers. The nutritional benefits of probiotic lactic acid bacteria contribute to increase consumption of minimally processed vegetables enriched with these microorganisms in supermarkets and restaurants, since the modern consumer search products of high functionality and safety. The aim of this study was to assess the viability of *Lactobacillus rhamnosus* HN001 on minimally processed yellow melon and determine the microbiological and physicochemical properties of this food. The counts of *L. rhamnosus* were above 10<sup>8</sup> CFU g<sup>-1</sup>, and the microbiological quality of melons was safe to consumers. The pH lowered and the acidity increased over time in minimally processed melons. The soluble solids did not differ between samples. The color coordinates L\* and a\* have not changed and melon firmness decreased over time. The scanning electron microscopy revealed adhesion of *L. rhamnosus* HN001 on the surface of treated melon. Despite some physicochemical changes, the production of minimally processed melon enriched with *L. rhamnosus* is feasible transforming it into a potential vehicle for probiotics.

Key words: Fruits, minimally processed, Lactobacillus rhamnosus, functional food, non-dairy products

## Resumo

A demanda por uma alimentação saudável com alimentos frescos, especialmente frutas e hortaliças minimamente processadas, resultou em uma variedade de produtos disponíveis para os consumidores. Os benefícios nutricionais de bactérias láticas probióticas contribuem para aumentar o consumo de vegetais minimamente processados enriquecidos com estes micro-organismos em supermercados e restaurantes, uma vez que o consumidor moderno busca produtos de alta funcionalidade e segurança. O objetivo deste estudo foi avaliar a viabilidade de *Lactobacillus rhamnosus* HN001 em melão minimamente processado e determinar as propriedades microbiológicas e físico-químicas desse alimento. As contagens de *L. rhamnosus* estavam acima  $10^8$  UFC g<sup>-1</sup> e a qualidade microbiológica dos melões estava segura para os consumidores. O pH diminuiu e a acidez aumentou ao longo do período de estocagem dos melões minimamente processados. Os sólidos solúveis não diferiram entre as amostras. As coordenadas de cor L \* e a \* não mudaram e a firmeza do melão diminuiu ao longo do tempo. A microscopia eletrônica de varredura revelou a adesão de *L. rhamnosus* HN001 na superfície do melão tratado. Apesar de algumas alterações físico-químicas, a produção de melão minimamente processado enriquecido com *L. rhamnosus* é viável e transforma-o em um potencial veículo de probióticos.

Palavras-chave: Frutas minimamente processadas, L. rhamnosus, alimentos funcionais, produtos não lácteos

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### Introduction

Minimally processed (MP) fruits represent one of the most rapidly expanding segments of the lightly treated refrigerated food market owing to their increased functionality. Minimal processing offers consumers highly nutritious, convenient and healthful fruits while maintaining freshness of the non-processed products (WU; ZHANG; WANG, 2012). According to Silva et al. (2007), minimally processed fruits and vegetables are products that have undergone cleaning, washing, selection, peeling and cutting, to obtain a 100 % usable product that is packaged and maintained under refrigeration.

Melon is a kind of fruit having a big market share in minimally processed products (SUPAPVANICH; TUCKER, 2011). Besides, this fruit is cholesterol free and it contains vitamin B1, B2, PP, A and C, being a fruit widely produced and consumed worldwide (NICOLAIS et al., 2011).

There is a consensus among researchers that the consumption of adequate amounts of fruits and vegetables is a key factor in a healthy diet (OMS-OLIU; SOLIVA-FORTUNY, 2011). These products contain significant levels of functional nutrients, such as fiber, vitamins, minerals and bioactive compounds, which assist in improving an individual's health and nutritional status. Thus, the consumption of food enriched with physiologically active components, such as probiotics (MARTINS et al., 2013), has led the industry to develop alternative products capable of improving health and wellness (MARK-HERBERT, 2004).

Probiotic dairy products are well established and recognized by most consumers as healthy. However, since many people are allergic or intolerant to these products, the consumption of fruits containing probiotic bacteria could be an alternative for these consumers (RIVERA-ESPINOZA; GALLARDO-NAVARRO, 2010; MARTINS et al., 2013). In addition to providing probiotic microorganisms to the consumer, fruits with probiotic bacteria contain nutrients beneficial to health and may positively influence the viability of probiotics (YOON; WOODAMS; HANG, 2004; RANADHEERA; BAINES; ADAMS, 2010).

Lactic acid bacteria are considered challenging microorganisms due to the essential amino acids and vitamins requirements for their growth (SALMINEN; VON WRIGH, 1998). Rodgers (2008) related that adding lactic acid bacteria to restaurant meals, such as vegetable salad, is a novel concept for the general public and food service professionals. Thus, the nutritional benefits of probiotic bacteria have motivated the increased consumption of minimally processed vegetables in supermarkets and restaurants. Such benefits may be assigned to the control and stabilization of the intestinal microbiota after the use of antibiotics (SOCCOL et al., 2010), increased gastrointestinal resistance to colonization by pathogens (ZHANG et al., 2011), competition for nutrients and adhesion sites (FERREIRA, SILVA, 2010; SAAD; BEDANI; MAMIZUKA, 2011), among others.

Raw food products have recently been intensively investigated as potential substrates for the production of probiotic non-dairy foods (MARTINS et al., 2013; SOCCOL et al., 2010; CETIN, 2011; PERES et al., 2012; YU et al., 2012). Alegre et al. (2011) and Rößle et al. (2010a; 2010b) found that probiotic strains have the ability to grow in fruit matrices due to fruit nutritional content and internal tissue structures. Therefore, this study aimed to evaluate the viability of *Lactobacillus rhamnosus* HN001 in minimally processed yellow melon and to determine their microbiological and physicochemical characteristics during storage.

## **Material and Methods**

### Preparation of probiotic culture

The culture of *L. rhamnosus* HN001 was prepared according to Rößle et al. (2010a). Briefly, the probiotic culture was activated twice in Man Rogosa Sharpe (MRS) broth and incubated at 37 °C for 18 h. Afterwards, the probiotic culture was activated in MRS broth for 16 h, then centrifuged at 5 °C for 15 minutes at 7000 g. The supernatant of the culture medium was discarded and the probiotic cell pellet was aseptically resuspended in a buffer solution of citric acid:sodium citrate at a 1:1 ratio and pH 6.2 at a ratio of 1:10; i.e., for every gram of cells, 10 ml of buffer solution was added to obtain at least  $10^{10}$  cells·mL<sup>-1</sup>.

# *Minimal processing of melon and inoculation with L. rhamnosus*

The melons, yellow type (*Cucumis melo* L.), were acquired in the Rio Pomba market, Minas Gerais State, Brazil. They were initially washed with potable water and sanitized in chloride solution with 200 mg L<sup>-1</sup> of active choride for 15 min at 5 °C. After sanitizing, melon fruits were rinsed in chloride solution 20 mg L<sup>-1</sup> for 5 min. Then, they were peeled, all seeds were removed and the fruits were cut into cubes of approximately 2 cm x 2 cm.

The melon cubes after cutting were immersed for 10 min in a solution citric acid:sodium citrate at a 1:1 ratio and pH 6.2 containing approximately  $10^{10}$  CFU·mL<sup>-1</sup> of *L. rhamnosus* HN001. Thus, to obtain minimally processed melon containing probiotic culture, 1 ml of the previously prepared probiotic cell solution was added for each gram of melon cubes, i.e., the ratio was 1 mL of probiotic solution for each gram melon. Subsequently, minimally processed melons were drained for 3 minutes, in domestic drainer, to remove excess of *L. rhamnosus* HN001 solution. The control treatment was immersed for 10 min in a buffer solution of citric acid:sodium citrate at a 1:1 ratio and pH 6.2 without inoculation of *L. rhamnosus*.

Minimally processed melons inoculated with *L. rhamnosus* HN001 and control were packed in polyethylene tereftalato (PET) boxes and stored at 6 °C and 15 °C for 0, 48, 96 and 120 h to monitor shelf life. All experiments were performed in three replicates.

# Assessment of L. rhamnosus viability on minimally processed melon

The viability of *L. rhamnosus* HN001 on minimally processed melons was assessed, in triplicate, by colony counting using pour plate method in Rogosa Agar SL (HIMEDIA, India), a selective agar used for lactobacilli enumeration, after 0, 48, 96 and 120 h of storage at 6 °C and 15 °C. The Petri dishes were incubated in anaerobic jars at 37 °C for 72 h.

#### Determination of the microbiological characteristics

Evaluations of fecal coliforms and *Salmonella* sp. are required by the National Agency of Sanitary Surveillance (ANVISA) as part of the Brazilian Health Ministry in the legal resolution RDC No. 12 (BRASIL, 2001). Based on these microbiological requirements, the Most Probable Number (MPN) method was used to determine fecal coliforms per gram of minimally processed melon (KORNACKI; JOHNSON, 2001). Also, analysis of *Salmonella* sp. in 25 g of sample from all treatments was done according to Andrews et al. (2001). All analyzes were carried out in triplicate.

#### Determination of physicochemical characteristics

Analyses of pH, acidity in citric acid and total soluble solids (TSS) in the minimally processed melons inoculated with *L. rhamnosus* HN001 and of the control treatment were conducted, in triplicate, according to AOAC (AOAC, 1997).

#### Colorimetric analysis

The surface color of minimally processed melons inoculated with *L. rhamnosus* HN001 and control was evaluated, in triplicate, using the colorimeter MiniScan EZ System (HunterLab, Reston, VA). Colorimetric analysis was performed by direct reading in reflectance mode of the L\*, a\*, b\* coordinates using the CIELAB L\* scale, as this scale has been adopted as the standard by the International Commission on Illumination. Three readings were done for each specimen, taken at different points of the fruit sample in order to obtain an average value.

#### Firmness analysis

The firmness of minimally processed melons inoculated with *L. rhamnosus* HN001 and of the melons of the control treatment was determined in triplicate by compression test with 5 melon cubes using a texturometer (CT3/BrookField). A fruit sample was placed on a flat surface and compressed for 1 second with a cylindrical probe (diameter of 5 cm). The fruits were analyzed at a distance of 5 mm, with a compression speed of  $5 \cdot \text{mm s}^{-1}$ .

#### Scanning electron microscopy (SEM)

The minimally processed melons inoculated with *L. rhamnosus* HN001 and the melons of the control treatment were analyzed with the Scanning Electron Microscope (SEM, model Zeiss LEO 1430 VP, Cambridge, England) immediately after the minimal processing (0 h) and after 120 h of storage at 6 °C to evaluate the adhesion of the cell to the fruit tissue. Analyses were performed in triplicate.

Fruit samples were sliced into sections of 0.5 cm x 0.5 cm with approximately 1-2 mm of thickness. Slices of fruit were fixed with a mixture of 5 % glutaraldehyde and a sodium phosphate buffered (PBS) solution (0.1 mol·L<sup>-1</sup>) at a ratio of 1:1 for 18 h at 7 °C. Samples were washed with PBS buffer 0.05 mol L<sup>-1</sup> (pH 7.2) for 10 min. Samples were dehydrated with a series of acetone treatments at concentrations of 30, 50, 70 and 90 °GL for 10 min each, then treated three times in acetone at 100 °GL for 10 min. Fruit samples were transferred to the critical point dryer (CPD020 model, Balzers Liechstenstein) for total sample dehydration. Dried samples were metalized using an FDU 010 metalizer

(Bal-Tec, Balzers Liechstenstein) for further SEM observation and images analysis in field 6.

#### Statistical analysis

Data were analyzed by completely randomized design in sub-plots divided by treatments, time and temperature. The data was analyzed using the Tukey test and regression analysis at 5 % of probability. The statistical analyses were carried out using the STATISTICA 7.0 software–(StatSoft, Inc., Tulsa, Okla., U.S.A.).

#### **Results and Discussion**

## Viability of L. rhamnosus in minimally processed melon

Lactobacillus rhamnosus HN001 was found to be above 10<sup>8</sup> CFU·g<sup>-1</sup> in minimally processed melons stored at 6 °C and 15 °C during the storage time and no significant difference was observed over time at different temperatures (Table 1). The efficacy of addition of probiotic bacteria in foods depends on the cell concentration of the inoculum and their viability that must be maintained during the shelf life of the product independent of food matrix (BERNARDEAU et al., 2008). To observe a beneficial effect in humans, the population of viable probiotic bacteria should range 106-1010. CFU·mL-1 or g-1 (GIALAMAS et al., 2010). The recommendation of the National Agency of Sanitary Surveillance (ANVISA) is an intake of a minimum of 108 to 109 CFU·day-1 of viable microorganisms (BRASIL, 2008). Thus, minimally processed melon can be a new alternative as the carrier of probiotic microorganisms. Therefore, a package containing 100 g of minimally processed probiotic melon offers the consumer a population above 10<sup>10</sup> CFU·g<sup>-1</sup>, sufficient quantity of bacteria to promote benefits to the host organism (DAVE; SHAH, 1997; SAAD, 2006).

Time (h) –	Counts of minimally processed melon inoculated with L. rhamnosus Log (CFU·g <sup>-1</sup> )			
	6 °C	15 °C		
0	$9.12\pm0.38^{\mathrm{aA}}$	$9.12\pm0.38^{\mathrm{aA}}$		
48	$8.90\pm0.05^{\rm aA}$	$9.10\pm0.20^{\mathrm{aA}}$		
96	$8.94\pm0.09^{\rm aA}$	$9.13\pm0.25^{\mathrm{aA}}$		
120	$8.74\pm0.18^{\mathrm{aA}}$	$8.74\pm0.09^{\mathrm{aA}}$		

**Table 1.** Viability of *L. rhamnosus* HN 001 (n = 3) in minimally processed melon.

CFU = Colony Forming Units. Different lowercase letters mean significant difference (p<0.05) between the temperatures and different capital letters mean significant difference (p<0.05) between the times. **Source:** Elaboration of the authors.

Rößle et al. (2010a) used *L. rhamnosus* GG in minimally processed apples and found that after 10 days of storage the product contained  $10^8$  CFU·g<sup>-1</sup> of *L. rhamnosus*, indicating the potential use of probiotic cultures in products of vegetal origin. Furthermore, Rößle et al. (2010b) used this lactobacilli strain on minimally processed apple added with oligofructose and inulin to obtain a potentially symbiotic product. They found that minimally processed apple contained approximately  $10^7$  CFU·g<sup>-1</sup> of *L. rhamnosus* GG after 14 days of storage, which is enough to exert a beneficial effect on the host organism, compared to the results obtained for probiotic dairy products.

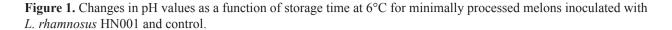
#### Determination of microbiological characteristics

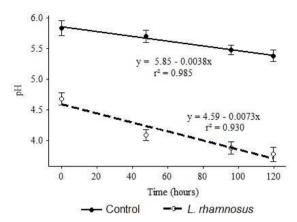
The resolution RDC n°. 12 by ANVISA (BRASIL, 2001) set as standard the maximum counting of fecal coliforms at 5.0 x 10<sup>2</sup> MPN per gram of fruit and the absence of *Salmonella* spp. in 25 g of sample. Thus, according to Brazilian legislation, the minimally processed melon inoculated with *L. rhamnosus* met the established microbiological requirements, with the results of fecal coliforms being less than 0.3 MPN·g<sup>-1</sup> after 120 hours of storage at 6 °C and 15 °C. However, the control treatment was improper for human consumption after 96 h storage at 15 °C, with a count of fecal coliforms above 1100 MPN·g<sup>-1</sup>,

as this value exceeds the limit set by Brazilian law. This result showed the importance of storage temperatures for the preservation of minimally processed vegetables. In addition to the action of lactic acid bacteria in preventing the growth and activity of undesirable microorganisms due to its great diversity of mechanisms of action, such as the production of organic acids, bacteriocins, hydrogen peroxide, diacetyl, among other compounds (PADMAJA et al., 2011). *Salmonella* sp. was absent in 25 g of all samples.

#### Determination of Physicochemical characteristics

The pH of melons was determined temperature of 21 °C. The lowest pH values for probiotic melons were due to live probiotic cells which multiplied and produced acid in 10 mL of buffer solution during the adhesion step to the initial evaluation (Time 0h). The pH value decreased in both minimally processed melon inoculated with *L. rhamnosus* and control samples during the storage time at 6 and 15 °C (p < 0.05) with no differences between the temperatures (Figure 1). However, differences in this parameter were not observed by Rößle et al. (2010a) in minimally processed apples containing *L. rhamnosus* GG after 10 days of storage at 2 and 4 °C.



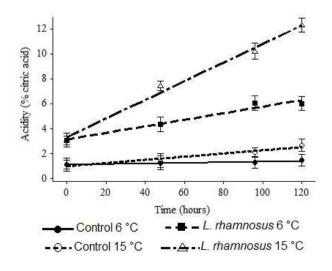


Source: Elaboration of the authors.

The acidity increased as a function of storage time for the control treatment stored at 15 °C and for minimally processed melons inoculated with *L. rhamnosus* stored at 6 and 15 °C (p < 0.05) (Figure 2). This occurred probably due to fruit sugar degradation and consequent acid release. Alegre et al. (2011) worked with minimally processed apples inoculated with *L. rhamnosus* and observed differences in total acidity between control and apples inoculated with this bacterium after 7 days of storage at 10 °C.

However, there were no significant differences regarding total soluble solids (p > 0.05) among treatments after 120 h of storage at both temperatures (control 6 °C: 10.01 ± 2.16 °Brix; control 15 °C: 9.63 ± 1.94 °Brix; Melon with *L. rhamnosus* 6 °C: 9.23 ± 1.45 °Brix; Melon with *L. rhamnosus* 15 °C: 9.59 ± 1.66 °Brix). Rößle et al. (2010a) also found no significant differences in total soluble solids content in minimally processed apples inoculated with *L. rhamnosus* and control treatment during storage.

**Figure 2.** Changes in acidity values as a function of storage time at 6 and 15 °C for minimally processed melon inoculated with *L. rhamnosus* HN001 and control.



Source: Elaboration of the authors.

#### Melon colour

The L\* coordinate, which represents the luminosity, was not affected (p > 0.05) by storage time and temperature (Table 2). However, minimally processed melon inoculated with *L. rhamnosus* presented higher average values of this parameter (lighter color) compared to control treatment. The a\* coordinate, which measures the hue or color type, was not affected (p > 0.05) by storage time or temperature (Table 2). However, the b\* coordinate, which represents saturation or color purity, differed significantly among treatments after 96 and 120 h (p < 0.05). The melons inoculated with probiotic bacterial culture presented lower average values when compared with the control

treatment (Table 2). In other words, the control melons showed the closest color to yellow. These results can be explained by the use of sodium citrate:citric acid buffer. This solution was used as a probiotic carrier for inoculating melon samples, and previous research indicates that citric acid can inhibit fruit browning in minimally processed fruit (ROJAS-GRAÜ; SOLIVA-FORTUNY; MARTIN-BELLOSO, 2009). Although control treatment has used same buffer solution, the sodium citrate:citric acid buffer solution containing the probiotic microorganism was more viscous and during the draining step it was possible that greater amount of citric acid may have been adhered in the product, reducing the browning of the fruit.

Treatment	Time (h)					
Treatment	0	48	96	120	Regressior	
		L* coordi	nate (6 °C)			
Control melon	66.06±0.38 <sup>b</sup>	68.18±0.52 <sup>b</sup>	65.33±0.32 <sup>b</sup>	$66.47 \pm 0.27^{b}$	NS	
Melon with L. rhamnosus	$68.19 \pm 0.48^{a}$	70.69±0.21ª	$67.92{\pm}0.46^{a}$	$69.85{\pm}0.57^{a}$	NS	
	L* coordinate (15°C)					
Control melon	66.06±0.24 <sup>b</sup>	67.55±0.65 <sup>b</sup>	61.30±0.21b	65.29±0.62 <sup>b</sup>	NS	
Melon with L. rhamnosus	68.19±0.65ª	$70.24{\pm}0.68^{a}$	$68.08{\pm}0.58^{a}$	69.92±0.36ª	NS	
	a* <i>coordinate</i> (6 °C)					
Control melon	-4.54±0.58ª	-3.85±0.32ª	-3.78±0.68ª	-3.87±0.46ª	NS	
Melon with L. rhamnosus	-4.03±0.39ª	-3.97±0.42ª	-2.92±0.53ª	-3.43±0.57ª	NS	
		a* <i>coordin</i>	<i>tate</i> (15°C)			
Control melon	-4.54±0.35ª	-3.59±0.53ª	-2.40±0.32ª	-2.57±0.67ª	NS	
Melon with L. rhamnosus	-4.03±0.56 <sup>a</sup>	-3.97±0.32ª	$-2.92\pm0.59^{a}$	-3.43±0.78ª	NS	
	b* <i>coordinate</i> (6 °C)					
Control melon	21.69±0.82ª	19.10±0.26ª	21.39±0.35ª	20.68±0.41ª	NS	
Melon with L. rhamnosus	20.24±0.68ª	19.46±0.42ª	19.53±0.38b	19.34±0.33 <sup>b</sup>	NS	
	b* <i>coordinate</i> (15°C)					
Control melon	21.69±0.66ª	22.30±1.35ª	22.17±0.73ª	23.17±1.18 <sup>a</sup>	NS	
Melon with L. rhamnosus	20.24±0.85ª	20.09±0.98ª	16.47±0.69b	17.56±0.93 <sup>b</sup>	NS	

Means followed by same letter in each column have no statistical difference by Tukey test at 5 % probability. NS: not significant. **Source:** Elaboration of the authors.

#### Melon firmness

A significant difference regarding fruit firmness was observed among treatments stored at 6 and 15 °C, with a linear reduction occurring over time,

from 0 to 120 h (p < 0.05) (Table 3). At 6 °C, the firmness decreased from 32.41 to 18.89 N for the control treatment, while the firmness of minimally processed melon inoculated with *L. rhamnosus* decreased from 19.31 to 16.18 N. On the other

hand, the control melons stored at 15  $^{\circ}$ C presented a decrease in firmness from 32.4 to 5.9 N, while the

firmness of minimally processed melon inoculated with *L. rhamnosus* decreased from 19.3 to 7.1 N.

	Firmness (N)						
	Melon with I	L. rhamnosus	Control	melon			
Time (h)	6 °C	15 °C	6 °C	15 °C			
0	$19.31\pm3.21^{\mathrm{aB}}$	$19.3\pm5.21^{\mathrm{aB}}$	$32.41\pm5.24^{\mathrm{aA}}$	$32.4\pm4.25^{\mathrm{aA}}$			
48	$18.06\pm4.15^{abB}$	$18.64\pm5.03^{\mathrm{aB}}$	$28.96\pm7.59^{\mathrm{aA}}$	$18.66\pm4.89^{\mathrm{bB}}$			
96	$15.33\pm3.73^{\mathrm{bA}}$	$10.65\pm4.51^{\mathrm{bB}}$	$17.53 \pm 2.66^{\text{bA}}$	$5.9\pm4.66^{\rm cC}$			
120	$16.18\pm2.67^{\mathrm{bA}}$	$7.1\pm4.69^{\mathrm{bB}}$	$18.89\pm6.57^{\mathrm{bA}}$	$5.9 \pm 5.73^{\text{cB}}$			

**Table 3.** Changes in firmness values as a function of storage time for minimally processed melons inoculated with L.*rhamnosus* HN001 and control.

Different lowercase letters mean significant difference (p<0.05) between the times and different capital letters mean significant difference (p<0.05) between the treatments.

**Source:** Elaboration of the authors.

The reduction of firmness was an important factor affecting quality of fresh-cut yelow melon fruit during storage. In this study, we observed that after 5 days of storage at 15 °C, minimally processed melon with L. rhamnosus had no firmness acceptable for consumption. This can be explained by the action of probiotic bacteria and also the activity of pectinases and polygalacturonases. Thus the action of these enzymes and also the increase in the rate of metabolic processes leads to a limitation on shelf life of the vegetables. Furthermore, the addition of probiotic microorganisms potentiates these effects by the action of microbial enzymes. Therefore, it is suggested the use of alternative / technologies to improve the firmness and prolong the shelf life of these products, such as application of calcium salts, edible coatings, modified atmosphere, among others.

Similar to these results, Rößle et al. (2010a) found that minimally processed apples inoculated with *L*. *rhamnous* lost their firmness after the second day of storage at 5 °C. They pointed out that this result may be due to the immersion of apple slices in buffer solution containing the probiotic strain, which could induce tissue softening. Supapvanich and Tucker (2011) investigated the cell wall hydrolases in freshcut Honeydew melon fruit during storage at  $4 \pm 1$  °C for 5 days. They verified that firmness is a key factor affecting quality of minimally processed Honeydew melon fruit and associates with the increase in polygalacturonase and galactanase activities.

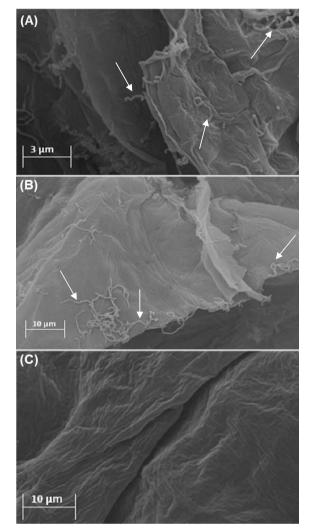
### *Observation of L. rhamnosus in minimally processed melon by Scanning Electron Microscopy (SEM)*

SEM analysis revealed the presence of several rod-shaped bacteria adhered to the surfaces of minimally processed melon inoculated with *L. rhamnosus* HN001 immediately after processing (Figure 3A) and after 120 h of storage at 6 °C (Figure 3B). This results suggest that melon is a promissory substrate for *L. rhamnosus*. No bacterial cell was found in the control treatment (Figure 3C).

Similar to our results, Martins et al. (2013) observed that fruits, such as apple, guava and banana have potential as carriers for probiotic bacteria. The results of scanning electron microscopy showed a positive interaction between the probiotic microorganisms and the fruity tissues, since bacteria strongly adhered to the fruit surface.

According to Oliveira et al. (2011), some steps of minimal processing, such as peeling and cutting, promote the release of intracellular content which is rich in minerals, sugars, vitamins and other nutrients, thereby creating ideal conditions for microbial growth. According to Rößle et al. (2010a) this allows the production of vegetal food as a carrier of probiotic microorganisms. Furthermore, according to Soccol et al. (2010), processed fruits and vegetables are considered good food matrices and provide an ideal substrate for probiotic cultures, since they contain minerals, vitamins, antioxidants and fibers.

**Figure 3.** SEM photomicrograph of minimally processed melon inoculated with *L. rhamnosus* HN001 (A) immediately after inoculation (0 h), (B) after 120 h of storage at 6 °C and (C) without the addition of *L. rhamnosus* HN001 (control treatment) immediately after processing stored at 6 °C. The arrows indicate that rod-shaped bacteria adhered to the vegetal tissue.



Source: Elaboration of the authors.

#### Conclusion

The production of minimally processed melon enriched with *L. rhamnosus* is feasible, since the probiotic bacterial count was maintained at high levels over the shelf life of the melons. Thus, this fruit has potential for use as a carrier of probiotic bacteria and constitutes an alternative for vegetarians and individuals who have restricted cholesterol diets, lactose intolerance or are allergic to milk proteins present in dairy products. Our previous research has shown that tropical fruits, such as guava, banana and papaya, as well as yacón roots, provide a good substrate for probiotic strains, presenting good adhesion to vegetal tissues.

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