

Postharvest quality of 'Prata Anã' bananas treated with microalgae coating

ABSTRACT

Aims: Biodegradable technologies to maintain the food quality without harming consumer health and the environment comprise an innovative tool in the post-harvest treatment of fruits. This study tests the efficiency of microalgae as a coating for bananas of the 'Prata Anã' cultivar.

Study design: The experimental design was completely randomized, with plots subdivided over time.

Place and Duration of Study: The experiment was conducted in Laboratory of Fruit Postharvest Technology of the Federal University of Campina Grande (UFCG), Pombal, Paraíba, Brazil.

Methodology: We applied the following treatments: 0% (no coating); 2% of *Chlorella* sp.; 2% of *Scenedesmus* sp.; and 2% *Spirulina platensis*. After application of treatments, the fruits were stored at 25 ± 2 °C and $65 \pm 5\%$ RH and analyzed every two days for 10 days.

Results: The bananas presented significant differences between the treatments at sixth and eighth days of storage, from the evolution in the color of the bananas, showing increase of the parameter a^* and reduction in the brightness and hue angle for both the bark and the pulp. The fruits covered with *Chlorella* sp. were presented at sixth days with mass loss of 5.89%, greater firmness of 65.24 N, besides 0.60% of malic acid and soluble solids, SS / TA ratio and total sugars of 25.50%, 21.73 and 8.27%, respectively.

Conclusion: The microalgae *Spirulina platensis*, *Chlorella* sp., and *Scenedesmus* sp. used at 2% in the coating of 'Prata Anã' bananas, delayed ripening, maintained pulp firmness, and decreased fresh mass loss, extending fruit conservation to eight days. Untreated fruits ripened fast, lost significant fresh weight, and softened.

Keywords: *Musa acuminata*; conservation; edible films; alimentary safety.

1. INTRODUCTION

The banana (*Musa spp.*) is one of the main fruits produced worldwide, being one of the most important tropical fruits due to its pleasant flavor, high nutritional properties, and natural fruiting throughout the year [1]. In Brazil, bananas are the second most produced fruits, losing only for oranges. The total Brazilian production was 6.7 million tons in 2016 [2], with the 'Prata Anã' cultivar leading the production, which shows its good acceptance among consumer.

Recent studies show the increase of fruit shelf life with the use of edible coatings, which can delay the metabolism of fruits and increase their useful life. Edible coatings can be used to inhibit the exchanges of humidity, oxygen, and carbon dioxide, improving the characteristics of covered products [3].

The choice of coating raw material comprises a crucial step to improve the results of coatings. The microalgae stand out among the several materials studied for coating elaboration with the objective of extending the useful life of fruits and vegetables.

The *Spirulina platensis* stands out among the species of microalgae that can be used in fruit coating due to its high amount of proteins (64 to 74%), vitamins (A, B, B2, B6, B12, E, and D), minerals, carbohydrates, carotenoids, beta-carotene, xanthophyll, γ -linolenic acid, and antioxidant activity [4, 5, 6]. A study using *S. platensis* coating in the concentration of 3% associated with 3% of cornstarch showed an increase of 30% in soluble solids, 15 N of pulp firmness, of fresh mass loss less than 3.5%, and substantial vitamin C content in mangos 'Tommy Atkins' stored for 11 days at 10 °C and 63% RH [7] and one day under ambient conditions was added to each interval (21.2 ± 0.5 °C and $51 \pm 2\%$ RH).

Besides the *S. platensis*, the *Chlorella sp.* also has a rich content of proteins, fatty acids, starch, minerals, phycocyanin, chlorophyll, beta-carotene, astaxanthin, vitamins, lipids, enzymes, and essential amino acids [8]. The proteins of the *Chlorella sp.* have a high emulsifying capacity compared with other ingredients [9]. This rich composition brings several benefits to the human and animal health, as well as providing a protective activity due to the presence of butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) antioxidants [10]. Such compounds add value to the products making a promising raw material for fruit coating.

The *Scenedesmus sp.* is used in many studies for industrial application, such as for the production of biodiesel, bioethanol, polymers, and food additives due to its ease of cultivation and the capacity to survive in extreme environments. This microalgae, depending on the cultivation technique, contains lipids (up to 60%), proteins (up to 60%), polysaccharides (up to 50%), and high value compounds, such as astaxanthin and lutein, besides containing all essential amino acids and high amounts of macro and microelements [11, 12].

In this sense, this work aimed to assess the potential of the *Spirulina platensis*, *Chlorella sp.*, and *Scenedesmus sp.* microalgae for the use as a coating in 'Prata Anã' bananas analyzed periodically under storage conditions of room temperature.

2. MATERIAL AND METHODS

Bananas of the 'Prata Anã' cultivar were obtained in Petrolina, Pernambuco, Brazil. The bunches were harvested at the stage of maturation 2 (green peel with yellow lines), according to Brazilian banana classification standards [13]. The fruits were packed in single layers in containers previously covered with shredded paper to minimize the impact and friction and then transported to the Laboratory of Fruit Postharvest Technology of the Federal University of Campina Grande (UFCG), Pombal, Paraíba, Brazil. In the laboratory, we selected the fruits for uniform size and color, discarding those with defects or injuries due to transport. The fruits were washed with 1% solution of neutral detergent and, after rinsing, sanitized with sodium hypochlorite solution at 100 mg L⁻¹ for 15 minutes before outdoor drying.

The experimental design was completely randomized, with plots subdivided over time with four repetitions and three fruits per plot. The sampling over time occurred every two days during 10 days of storage at room temperature (25 °C and 65% RH). We applied the following treatments: T1 – 0% (control, without coating); T2 – 2% of *Chlorella sp.*; T3 – 2% of *Scenedesmus sp.*; T4 – 2% of *Spirulina platensis*. The concentrations were established according to the results of previous studies [14, 15].

The microalgae used in this study were produced in tanks of organic production, in the Tamanduá Farm, Patos, Paraíba [16]. After the harvest, the coating concentrations were obtained by diluting solutions containing 2% biomasses of *Chlorella sp.*, *Scenedesmus sp.* and *S. platensis* in 2 L of distilled water under constant shaking until the homogenization. The treatments were applied through the immersion of fruits in the solutions. We analyzed the following variables: The color was assessed with a chroma meter (Konica Minolta, CR-400) through two readings in the equatorial area of the fruit and two readings in the pulp through transverse cut, being used the color space L*a*b* for reflectance. The measured color parameters regarding to plate-pattern were: L*, a*, b*, the angle hue ($h^{\circ} = \arctang(a^*/b^*) (-1)+90$) for a* negative, and ($h^{\circ} = 90 - \arctang(a^*/b^*)$) for a* positive, and the saturation index or chromaticity ($C^* = [(a^*)^2 + (b^*)^2]^{1/2}$).

The firmness (N) was measured with a digital penetrometer (Instrutherm, model PTR-300). Measurements were made in two opposite points of the equatorial area of the fruit with peel using a ferrule of 6 mm [17]. Loss of fresh mass (%) was obtained by the variation in the mass of the fruits over time. The fruits were weighted on the first day of experiment and every two days until the end of storage.

The homogenized pulp used in the evaluations of titratable acidity, soluble solids, and total soluble sugars was obtained after removal of peels and processing in a blender. Titratable acidity (% malic acid) was measured through the titrating of 5 g of banana pulp diluted in 50 mL of distilled water and add up two drops of 1% phenolphthalein under constant agitation, with a solution of sodium

hydroxide at 0.1 M [18]. The pH was obtained by direct reading of homogenized pulp in a digital pH meter (Digimed DM-22) [18].

Soluble Solids (%) was measured through direct reading in digital refractometer (Instrutherm) [17]. The division between values of soluble solids and titratable acidity produced the SS/TA ratio. Total sugar (%) was obtained by the anthrone method [19] with modifications. A sample of 0.5 g of pulp was diluted in 100 mL of distilled water. From fruits with four days, we take aliquots of 500 µL adding 500 µL of distilled water totaling 1 mL. From fruits with six days onwards, we take aliquots of 300 µL adding 700 µL of distilled water totaling 1 mL. To each aliquot, we added 2 mL of anthrone a tube under agitation and immersed them in a water bath at 100 °C for 5 minutes. After cooling, the samples were taken to the spectrophotometer for reading at the wavelength of 620 nm.

The data were submitted to analyses of variance and Student's t-test to verify significant differences among treatments at 5% of probability. Polynomial regressions were used to assess the behavior of variables over time. The tests were in the software SISVAR version 5.6 [20].

3. RESULTS AND DISCUSSION

The results of the variance table can be seen in Tables 1 and 2. On the first day of the experiment a twelve fruits sample was used for evaluation of the color of peel and pulp, firmness (N), fresh mass loss, titratable acidity (TA), pH, soluble solids (SS), SS / TA ratio, and total sugar, demonstrating the characteristics that the bananas presented at the time of harvest (Table 3).

Brightness and chromaticity of the peel showed a quadratic behavior over the storage period, the banana peel color praticamente não variou for the brightness and chromaticity of the peel, 57.07 and 40.79, respectively, after 10 days of storage they showed an average of 57.27 for brightness and 41.09 for chromaticity (Figure 1A and B). However, it is possible to observe a significant difference between the sixth day coverings, in which the control presents values of -3.80 and the other coatings presenting mean values of -14.40, and at the eighth days in which the control fruits evolved to 6.95, and fruits with 2% of *Chlorella* sp., *Scenedesmus* sp. and *S. platensis* evolved to -8.06, -9.97, -11.27, respectively, in this way, there was a delay in the evolution of the bark of the coated bananas in relation to the non-coated ones, which presented an evolution in coloration of the faster shell (Figure 1F). During ripening, the banana shows a change of coloration from green peel to yellow, increasing brightness and chromaticity [21]. The use of other coatings such as pectin, egg albumen, carrageenan, gelatin, potato starch, xanthan, and cornstarch for the conservation of banana 'Caturra' cultivar (*Musa paradisiaca* L.) also shows a yellowish coloration during all storage period [22].

Table 1. Summary of the analysis of variance for (L*, h°, C* and a*) in peel and (L*, h°, C* and a*) in pulp in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5% RH.

FV	Medium Square								
	DF	L* peel	h° peel	C* peel	a* peel	L* pulp	h° pulp	C* pulp	a* pulp
Coating (R)	3	43.04 ^{ns}	86.73**	16.16 ^{ns}	34.63**	3.27 ^{ns}	5.09 ^{ns}	4.01 ^{ns}	1.02 ^{ns}
Residue 1	12	19.38	3.31	5.02	1.41	3.01	1.66	65.99	0.40
Time (T)	4	114.84**	3268.80**	17.58*	1559.09**	1094.71**	96.40**	85.34**	25.10**
R x T	12	24.66 ^{ns}	19.46**	5.74 ^{ns}	7.96**	11.67**	3.56*	8.09 ^{ns}	0.77*
Residue 2	48	16.97	7.36	3.68	3.08	2.79	1.73	5.22	0.39
CV (%)		7.44	2.17	5.06	12.19	2.20	1.46	8.39	5.06
Mean general		57.28	104.28	41.09	-9.73	77.44	88.94	27.58	41.09

Note. ns: not significant; ** significant at 1%, * significant at 5%.

Table 2. Summary of the analysis of variance for loss of fresh mass, pulp firmness, titratable acidity (TA), Soluble solids (SS), SS/TA ratio and total sugar (TS) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5% RH.

FV	Medium Square							
	DF	Mass loss	Firmness	TA	pH	SS	SS/TA	TS
Coating (R)	3	59.09**	294.46**	0.37**	0.45**	49.12**	493.50**	27.54**
Residue 1	12	2.78	12.57	0.02	0.03	5.51	49.74	0.32
Time (T)	4	407.40**	6334.77**	1.66**	5.03**	688.00**	146.52*	36.96**
R x T	12	2.80**	59.88**	0.32**	0.22**	28.46**	151.44**	20.00**
Residue 2	48	0.84	16.01	0.01	0.02	8.00	58.57	0.57
CV (%)		13.94	7.22	19.69	3.25	18.70	33.03	9.49
Mean general		9.27	53.20	0.73	5.00	13.83	21.35	6.99

Note. ns: not significant; ** significant at 1%; * significant at 5%.

Table 3. Initial Characterization of 'Prata Anã' bananas. Pombal-PB, 2018. (n=12)

Initial Characteristics	Averages ± SD
Fresh mass loss (g)	140.00 ± 11.05
Brightness in the peel (L*)	57.07 ± 2.45
Chromaticity in the peel (C*)	40.79 ± 1.08
Hue angle (h°) in the peel	116.58 ± 0.52
Brightness in the pulp (L*)	86.4 ± 0.34
Chromaticity in the pulp (C*)	21.43 ± 1.35
Hue angle (h°) in the pulp	92.08 ± 0.77
a* in the peel	-18.25 ± 0.66
a* in the pulp	-0.78 ± 0.30
Firmness (N)	63.65 ± 5.82
Titratable acidity (% malic acid)	0.33 ± 0.65
pH	5.73 ± 0.07
Soluble solids (%)	4.55 ± 0.63
SS/TA ratio	14.66 ± 0.52
Total sugar (%)	3.80 ± 0.29

The color angle h° of the peel decreased over time (Figure 1C), the fruits began with an average of 116.58°, characterizing a green coloration with yellow lines. After six days, the control had an angle (103.30°) lower than all treatments with coatings, while the coating with *Scenedesmus* sp. showed the largest value (111.43°). On the eighth day, *S. platensis* provided the largest value (105.88°), and again the smallest value was verified for the control fruits (95.21°). This reduction in the color angle agrees with the natural ripening of the fruit, occurring the change from green for yellow, indicating that at eight days of storage the uncovered fruits showed yellowish coloration and the covered ones still were yellow-greenish.

The brightness of the pulp decreased during the storage period (Figure 1D). All treatments showed similar behavior until the sixth day of storage, differing from the control on the eighth day, in which the control showed the smallest value (75.09). The coating to with *S. platensis* had the second smallest value (80.24), characterizing a darker pulp, which indicates a delay in the process of ripening of the pulp. With regard to the parameter a* in the pulp, it was verified that at sixth days the pulp of the bananas presented significant differences between the treatments, the control fruits presented values

of -0.40, while the recoils presented, respectively, 0.73, 0.81 and 0.42, *Chlorella* sp., *Scenedesmus* sp. and *S. platensis*. However at eighth days a great evolution in the coloration of the pulp of fruits covered with *S. platensis* and *Scenedesmus* sp. which presented the highest values of 0.86 and 0.85, respectively, followed by control fruits with 0.70. Indicating normal maturation evolution in fruit pulp coloration (Figure 1G).

The angle h° of the pulp had little variation over time (Figure 1E). The largest differences among coating types occurred in eight days of storage when the control showed the largest value (91.56°) and *S. platensis* the smallest value (87.95°). The control treatment showed the typical cream color of ripening banana pulp. The coated fruits showed yellow pulps with greenish lines, indicating a retard in ripening.

Previous studies show that increases in the concentration of coating with *Chlorella* sp. enhance the firmness and some changes in the color of peel in "Tommy Atkins" mangos [14]. Coatings with *Chlorella* sp. in the concentration 1% and 2% provides ripening without damage in the quality of fruits analyzed for 28 days of storage (21 days at 10°C and 42% UR and seven days at 25°C).

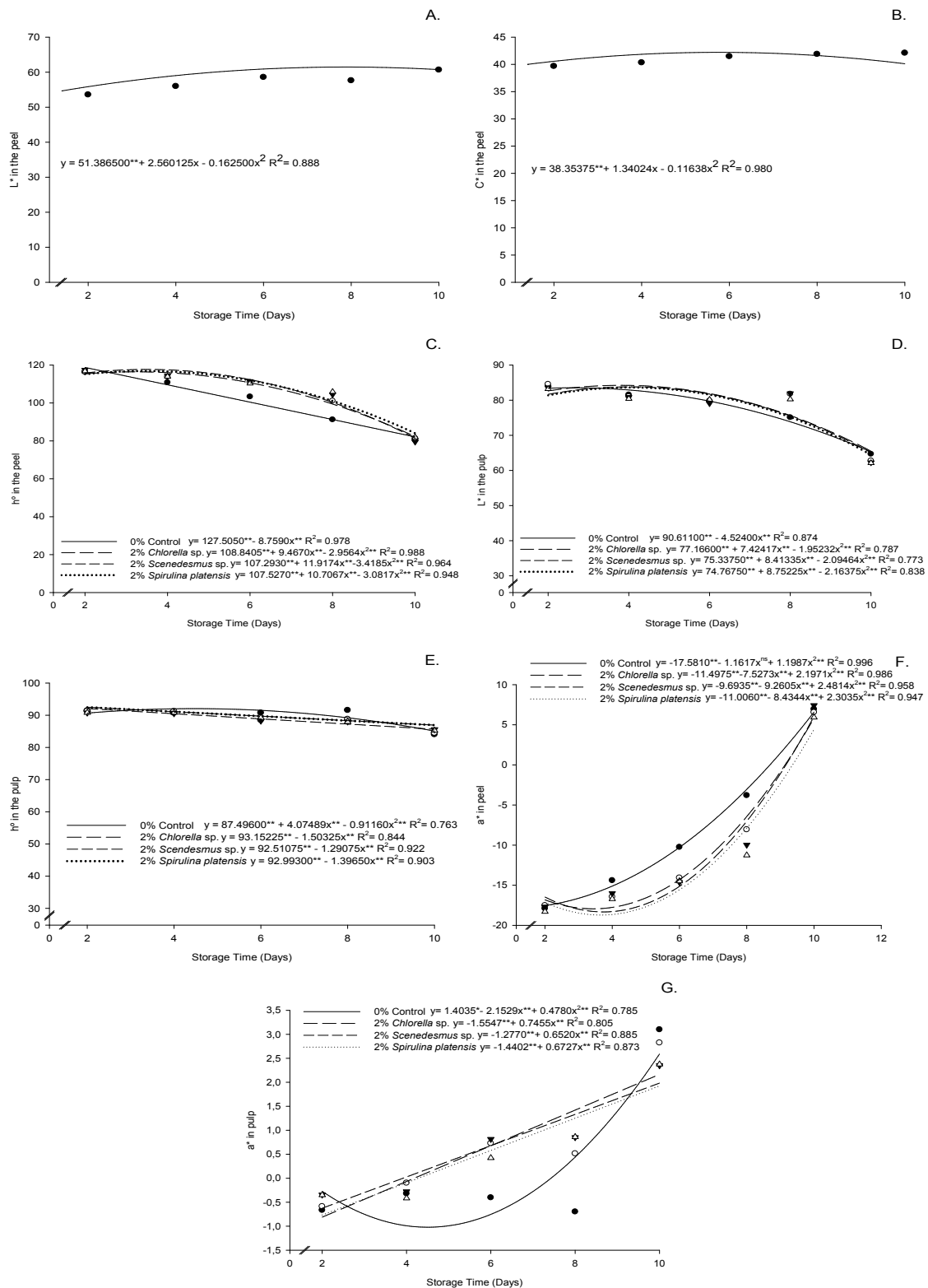


Fig. 1. Brightness of the peel (L *), chromaticity in the peel (C *) over the storage period in (A and B) hue angle in the peel (h°), brightness in the pulp (L *), hue angle in the pulp (h°), a* in the peel and pulp, respectively (C, D, E, F e G) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5% RH.

The loss of fresh mass increased with the storage time in all treatments (Figure 2). Fruits of the control treatment showed the largest mass loss. On the sixth day of storage, the control lost 10.12% of mass, while fruits coated with *Chlorella* sp. lost 5.89%, the smallest mass loss. In this way, the fruits covered with 2% of *Chlorella* sp. maintained turgidity until the eighth day. At the end experiment, the largest mass loss was verified in the controls fruits (18.82%), followed by *S. platensis* and *Scenedesmus* sp. (17.02 and 16.85%, respectively), and the smallest mass loss occurred in bananas covered with 2% of *Chlorella* sp. (14.81%).

Some studies conclude that the best indicators of physical-chemical maintenance of the quality in 'Tommy Atkins' mango were observed in fruits coated with 1% of *Chlorella* sp., as demonstrated in the excellent appearance, little loss of fresh mass, and high content of total sugars both at 10 °C and 23 °C [15].

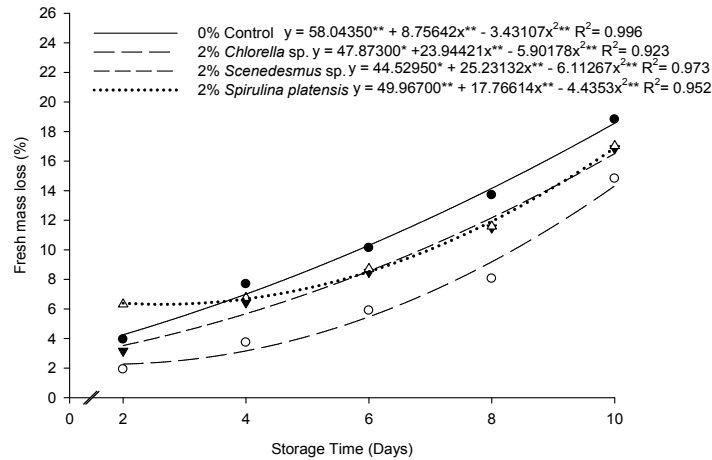


Fig. 2. Loss of fresh mass (%) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5% RH.

The **firmness** had a quadratic behavior, with a significant reduction at the end of storage in all treatments (Figure 3). On the sixth day, the highest **firmness** occurred in fruits coated with *Chlorella* sp. (65.24 N), while the control obtained the lowest value (53.61 N). At the end of storage (10 days), the largest firmness was observed in fruits with *S. platensis* (25.46 N), followed by of *Chlorella* sp. and *Scenedesmus* sp. with 16.01 N and 15.49, respectively, and the lowest firmness in the control treatment (15.35 N). Studies evaluating the effects of natural polymeric coatings in the conservation of 'Caturra' banana also detected reductions of **firmness** in all treatments over the storage, which indicates cell wall degradation and cell turgidity loss with ripening [22].

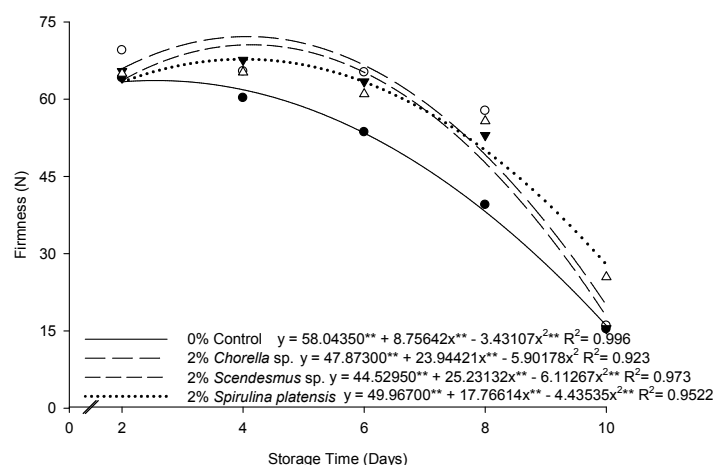


Fig. 3. Firmness (N) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at $25 \pm 2^\circ\text{C}$ and $65 \pm 5\%$ RH.

The titratable acidity increased in all of the studied treatments with the storage time (Figure 4A). There was no significant difference among the coatings until the sixth day of storage, in which the smallest values were observed in fruits with *Spirulina platensis* (0.45% of malic acid), followed by *Chlorella* sp. (0.60%), control, and *Scenedesmus* sp. (both 0.65%). At the end of storage, the control had the smallest values (0.72%) and *S. platensis* the highest values (1.33%). However, all the results obtained from the 8 days of storage were higher to the reported in other studies with banana of same variety, which range from 0.22% to 0.36% when stored for 35 days under cooling [23], and 0.2% to 0.5% for 'Prata' banana [24].

Due to the increasing tendency in the acidity, we verified a steady reduction in the pH values for all of the treatments (Figure 4B). The treatments showed significant differences in pH on the sixth and eighth days storage, with the smallest values occurring in control fruits and the ones coated with *Scenedesmus* sp. (4.41 and 4.14, respectively). The largest values were obtained by *S. platensis* (5.13), maintaining this result until the end of storage.

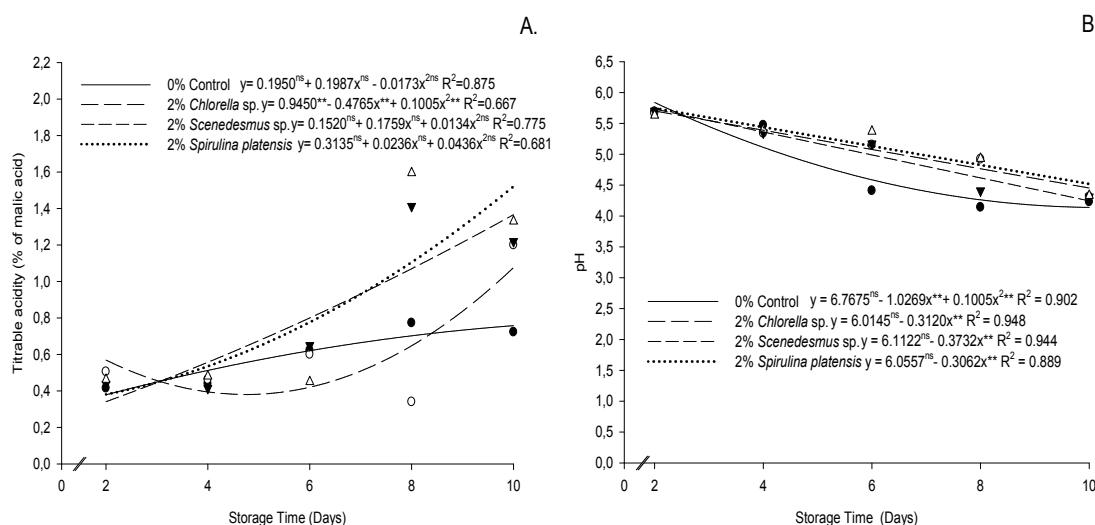


Fig. 4. Titratable acidity (% of malic acid) (A), and pH (B) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5% RH.

The soluble solids increased over time (Figure 5A). Until the eighth day of storage, the treatments with the largest values of soluble solids were the control with 20.95% followed by *Scenedesmus* sp. However, on the tenth day of storage, the coating of *Chlorella* sp. showed the largest values (25.50%) and the control fruits the smallest values (22.70%). This behavior of soluble solids suggests that the starch is being hydrolyzed in sugars through primary metabolism to supply breathing substrate for biological activities, with consequent increase of the sweetness [25].

In agreement with the increase of the acidity and soluble solids over the storage, the SS/TA ratio also increased over time, showing differences among treatments at the tenth day (Figure 5B). The control had the largest value (32.30), followed by *Chlorella* sp., *Scenedesmus* sp., and *S. platensis* (21.73, 21.58, and 17.66, respectively). A high SS/TA ratio is critical in the assessment of fruit flavor [26].

The **total sugar** showed different behaviors among treatments over the storage period (Figure 5C). Beginning with a value of 3.80%, the first high variation occurred at six days, in which the control had a pick of sugar synthesis of 11.70%, followed by *Chlorella* sp. (8.27%), *S. platensis* (4.10%), and *Scenedesmus* sp. (4.04%). At the end of storage, the fruits covered with *S. platensis* had the largest values (9.08%), followed by *Chlorella* sp. (8.52%), and low values in control fruits (6.82%) and *Scenedesmus* sp. (6.63%). Increases in total sugars are associated with the elevation of soluble solids contents although the measurement of soluble solids do not represent the exact content of sugars because they include other substances dissolved in vacuolar sap (vitamins, phenolics, pectins, and organic acids) [26]. However, the sugars are the most representative, comprising up to 85 - 90% of the soluble solids.

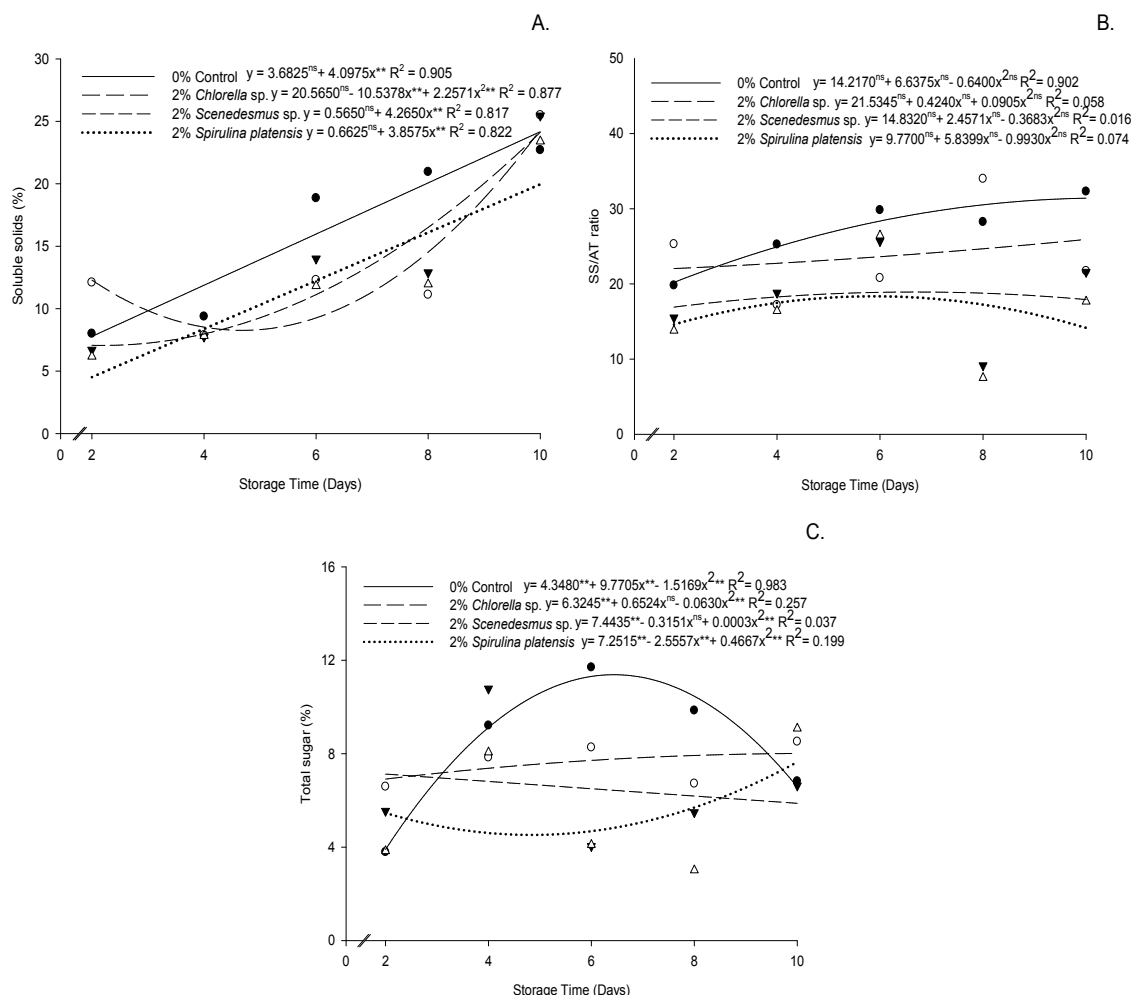


Fig. 5. Soluble solids (%) (A), SS/TA ratio (B), and total sugar (%) (C), in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at $25 \pm 2^\circ\text{C}$ and $65 \pm 5\%$ RH.

4. CONCLUSION

The microalgae *Spirulina platensis*, *Chlorella* sp., and *Scenedesmus* sp. used at 2% in the coating of 'Prata Anã' bananas, delayed ripening, maintained pulp firmness, and decreased fresh mass loss, extending fruit conservation to eight days. Untreated fruits ripened fast, lost significant fresh weight, and softened.

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