

Postharvest Quality of 'Prata Anã' Bananas Treated with Microalgae Coating

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Authors' contributions

This work was carried out in collaboration between all authors. Authors EFQF, JJFS, BGFLS and KAA performed the laboratory research, statistical analysis and preliminary sketch. While authors AMFO, AEMMT, EAO, MHBSR and LMO collaborated in the rewriting of the manuscript, improving the bibliographical revision with base in the obtained data. Authors RHCRA and JFL collaborated in the development of the study and they accomplished corrections of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

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

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Study Design: We carried out a completely randomized experimental design with plots subdivided over time.

Place and Duration of Study: The experiment occurred in the Laboratory of Fruit Postharvest Technology of the Federal University of Campina Grande (UFCG), Pombal, Paraíba, Brazil, during 10 days of storage.

Methodology: The following coatings were applied to the banana fruits: 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 ±5% RH and analysed every two days for 10 days.

Results: The characteristics of bananas differed among treatments at the sixth and eighth days of storage. The colour of bark and pulp showed an increase of the parameter a^* and reduction in brightness and hue angle over time. At the sixth day, fruits covered with *Chlorella* sp. showed 5.89% of mass loss of, firmness of 65.24 N, 0.60% of malic acid, 25.50% of soluble solids, 21.73 SS/TA ratio, and 8.27% of total sugars, showing retardation fruit ripening.

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.*), one of the most produced fruits worldwide, stands out among tropical fruits due to its pleasant flavor, high nutritional properties, and natural fruiting throughout the year [1]. Bananas are the second most produced fruits in Brazil, which had a total production of 6.7 million tons in 2016 [2]. The 'Prata Anã' cultivar leads the national production, showing 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 increasing their useful life. Edible coatings inhibit the exchanges of humidity, oxygen, and carbon dioxide, improving the characteristics of the products [3].

The choice of coating raw material comprises a crucial step to improve the results of coatings. The microalgae stand out among several studied materials. For example, the *Spirulina platensis* 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, a loss of fresh mass lesser than 3.5%, and substantial vitamin C content in 'Tommy Atkins' mangos stored for 11 days at 10°C and 63% RH [7], with one day under ambient conditions added to each interval (21.2 ±0.5°C and 51 ±2% RH).

The *Chlorella* sp. microalga 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 *Chlorella* sp. have a high emulsifying capacity compared with other ingredients [9]. This rich composition promotes 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. have several industrial applications, such as for the production of biodiesel, bioethanol, polymers, and food additives due to its ease 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 coatings in 'Prata Anã' bananas stored at room temperature conditions.

2. MATERIALS AND METHODS

Bananas of the 'Prata Anã' cultivar were obtained in Petrolina, Pernambuco, Brazil. The bunches

harvest occurred at the second stage of maturation (green peel with yellow lines), according to Brazilian banana classification standards [13]. The fruits were packed in single layers in containers previously filled with shredded paper to reduce the impact and friction during the transport 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 colour, discarding those with defects or injuries due to transport. The fruits were washed with 1% solution of neutral detergent and, after rinsing, sanitised with sodium hypochlorite solution at 100 mg L^{-1} for 15 minutes before outdoor drying.

The experiment comprised a completely randomized design 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). The following coatings were applied to the banana fruits: T1 – 0% (control, without coating); T2 – 2% of *Chlorella sp.*; T3 – 2% of *Scenedesmus sp.*; T4 – 2% of *Spirulina platensis*. The concentrations were chosen 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 2% concentrations were obtained by diluting 4.00 g the algal biomasses in 2 L of distilled water under constant shaking until the homogenisation. The fruits were immersed in these solutions to obtain the coatings of each treatment. After that, we assessed the changes in fruit characteristics over time measuring the variables below.

The colour 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 a transverse cut. We used the colour space $L^*a^*b^*$ for reflectance. The measured colour parameters regarding plate-pattern were: L^* , a^* , b^* , the hue angle ($h^\circ = \arctan(a^*/b^*)(-1)+90$) for a^* negative, and ($h^\circ = 90 - \arctan(a^*/b^*)$) 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 calculated by the variation in the mass of fruits over time. The fruits were weighted on the first day of experiment and every two days until the end of storage.

The homogenised 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 measurement of homogenised 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 took aliquots of 500 μL adding 500 μL of distilled water totaling 1 mL. From fruits with six days onwards, we took aliquots of 300 μL adding 700 μL of distilled water totaling 1 mL. To each aliquot, we added 2 mL of anthrone in 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 variations fruits characteristics over time. The tests were carried out 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. To verify the bananas characteristics at the time of harvest, on the first day of the experiment, a twelve fruits sample was used for evaluation of the colour of peel and pulp, firmness (N), fresh mass loss, titratable acidity (TA), pH, soluble solids (SS), SS/TA ratio, and total sugar (Table 3).

The brightness and chromaticity banana peel colour had only significant changes over time,

varying, respectively, from 57.07 and 40.79 at the beginning of the experiment to 57.27 and 41.09 after 10 days of storage (Fig. 1A and B).

However, the parameter a^* of the bark differed significantly among the treatments at the sixth day, in which the control showed values of -3.80 and the coatings an average of -14.40, and at the eighth days, in which the control fruits increased to 6.95, and fruits with *Chlorella* sp., *Scenedesmus* sp. and *S. platensis* increased to -8.06, -9.97, -11.27, respectively. These results suggest that the coating of bananas delayed the process of peel colour changes (Fig. 1F). During ripening, the banana shows a change of colouration 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 of the 'Caturra' cultivar (*Musa paradisiaca* L.) also showed a yellowish colouration during all storage period [22].

The Hue colour angle (h°) of the peel decreased over time (Fig. 1C). The fruits began with an average of 116.58° , characterising a green colouration with yellow lines. After six days, the Hue angle of the control was lower than all treatments with coatings (103.30°), while the coating with *Scenedesmus* sp. showed the highest value (111.43°). On the eighth day, *S. platensis* provided the highest value (105.88°), and again the smallest value was measured in the control fruits (95.21°). This reduction in the colour 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 colouration and the covered ones still were yellow-greenish.

The brightness of the pulp decreased during the storage period (Fig. 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 lowest value (75.09). The coating with *S. platensis* had the second smallest value (80.24), characterising a dark pulp, which indicates a delay in the process of ripening of the pulp. The a^* parameter in the pulp showed a significant difference among treatments on the sixth day. The control fruits had values of -0.40, while the coatings with *Chlorella* sp., *Scenedesmus* sp. and *S. platensis* showed a^* values of 0.73, 0.81, and 0.42, respectively. However, at eighth days, there was a substantial change in the colouration of pulp of

fruits covered with *S. platensis* and *Scenedesmus* sp., which had the highest values of 0.86 and 0.85, respectively, followed by control fruits with 0.70. Such results indicate a normal maturation evolution in the fruit pulp colouration (Fig. 1G).

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

Previous studies show that increases in the concentration of coating with *Chlorella* sp. enhance the firmness and promote some changes in the colour 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 analysed for 28 days of storage (21 days at 10°C and 42% UR and seven days at 25°C).

The loss of fresh mass increased with the storage time in all treatments (Fig. 2). Fruits of the control treatment showed the largest loss of mass. 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 of 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 *Chlorella* sp. (14.81%).

Studies state 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].

The curve of variation in firmness over time showed a quadratic behavior, with a significant reduction at the end of storage in all treatments (Fig. 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

Table 1. Summary of the analysis of variance for L*, h°, C*, and a* in peel and in pulp of 'Prata Anã' bananas submitted to different coatings with microalgae 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 Characterisation of 'Prata Anã' bananas. Pombal-PB, 2018. (n=12)

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

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].

The titrateable acidity increased in all of the studied treatments with the storage time (Fig. 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 lowest values (0.72%) and *S. platensis* the highest values (1.33%). However, all the results obtained from the 8 days of storage were higher than 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, the pH values decreased steadily in all treatments (Fig. 4B). The treatments showed significant differences in pH on the sixth and eighth day of 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.

The soluble solids increased over time (Fig. 5A). Until the eighth day of storage, the treatments with the largest contents 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 highest values (25.50%) and the control fruits the lowest values (22.70%). This pattern of variations in soluble solids suggests that the starch was hydrolysed 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 on the tenth day (Fig. 5B). The control had the highest 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 changes in total sugar over the storage period showed different patterns of variation among treatments (Fig. 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 highest values (9.08%), followed by *Chlorella* sp. (8.52%), and the lowest values were found in control fruits (6.82%) and *Scenedesmus* sp. (6.63%). Increases in total sugars are related with the elevation of soluble solids contents

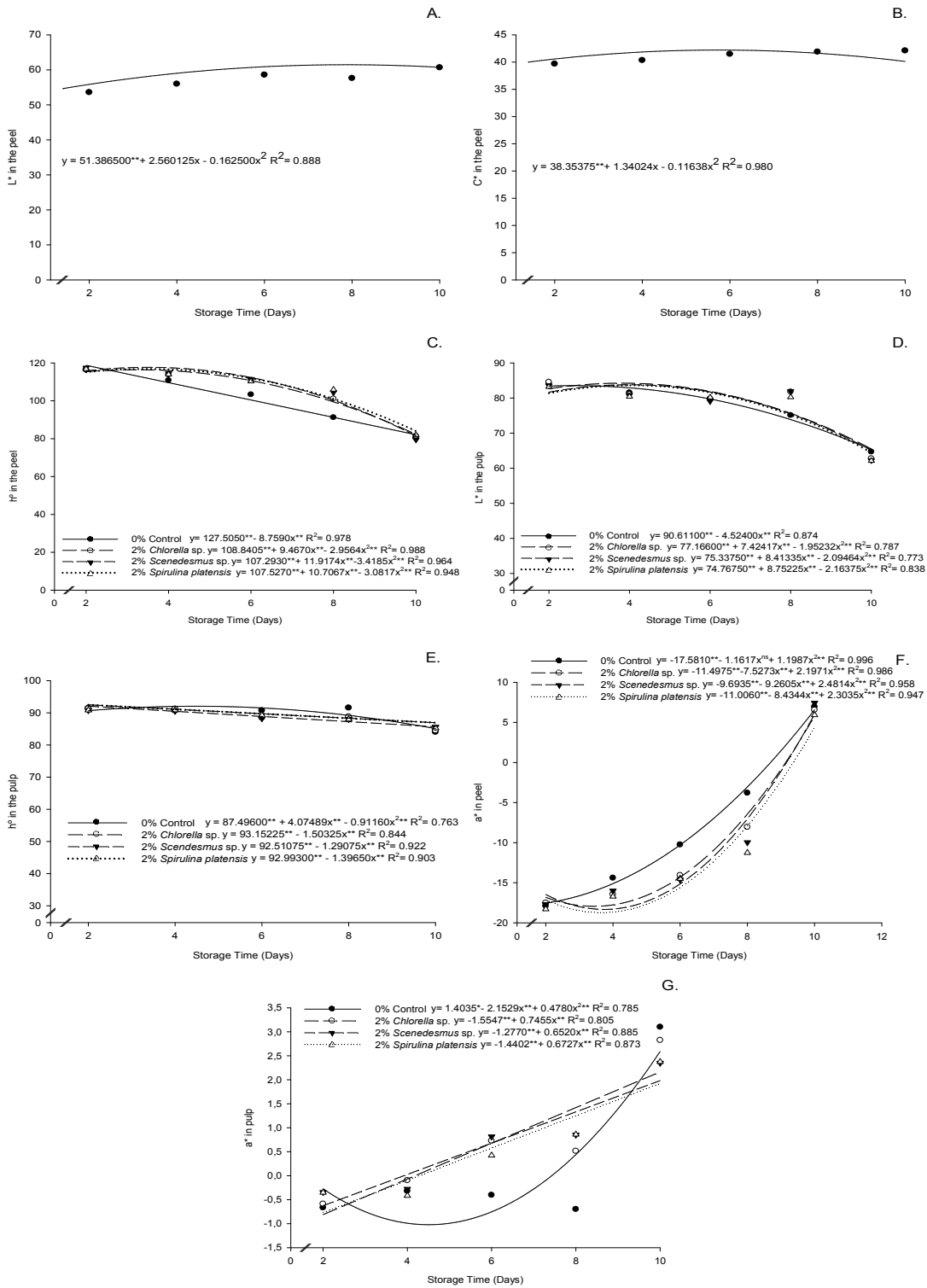


Fig. 1. Brightness of the peel (L^*), chromaticity in the peel (C^*), hue angle in the peel (h°), brightness in the pulp (L^*), hue angle in the pulp (h°), a^* in the peel, and a^* in the pulp (A, B, C, D, E, F, and G, respectively) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at $25 \pm 2^\circ\text{C}$ and $65 \pm 2\% \text{RH}$.

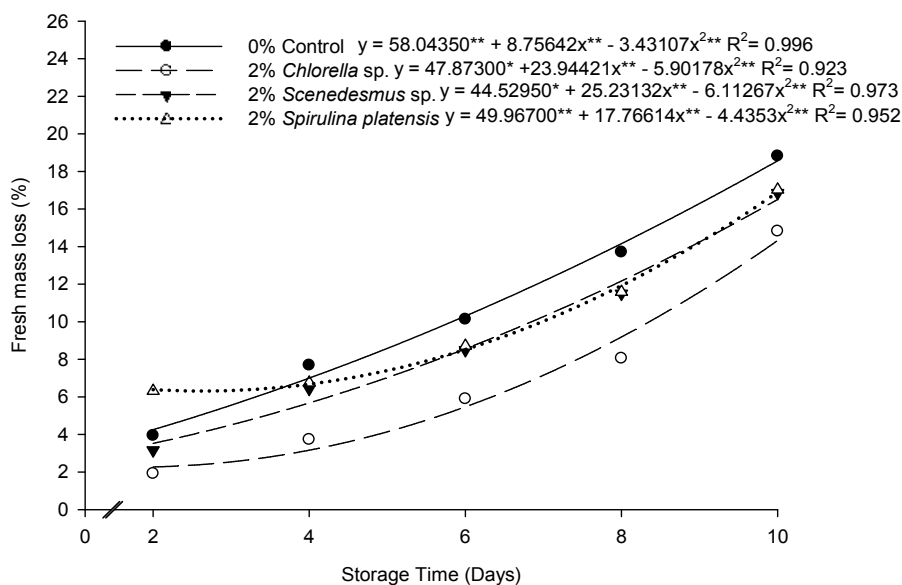


Fig. 2. Loss of fresh mass (%) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5 % RH.

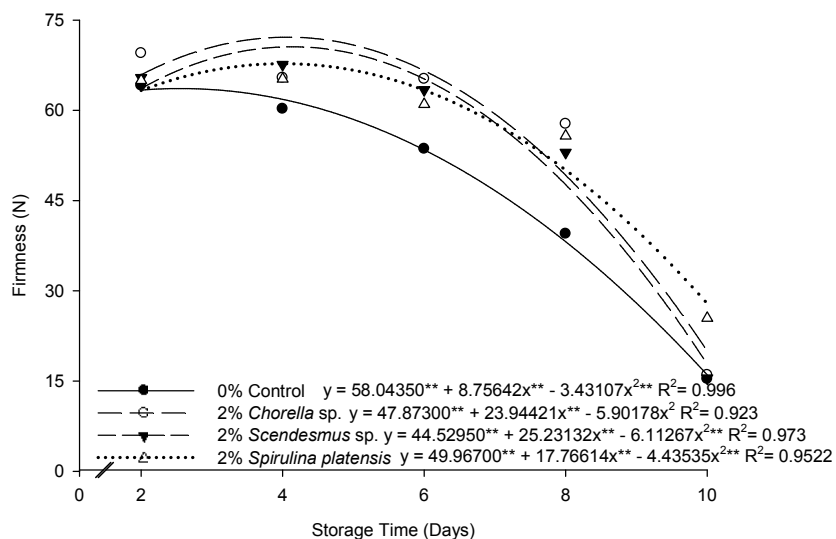


Fig. 3. Firmness (N) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at 25 ± 2 °C and 65 ± 5 % RH.

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. The variations in the carbohydrate contents of the fruits may have occurred due to small changes in the stages of banana maturation, causing random variations of the results found.

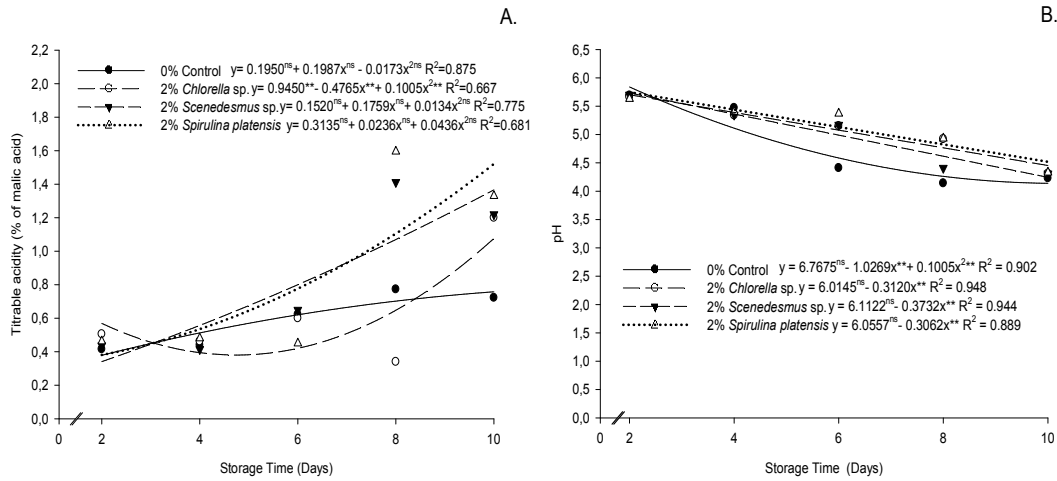


Fig. 4. Titratable acidity (% of malic acid) (A), and pH (B) in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at $25 \pm 2^\circ\text{C}$ and $65 \pm 5\%$ RH.

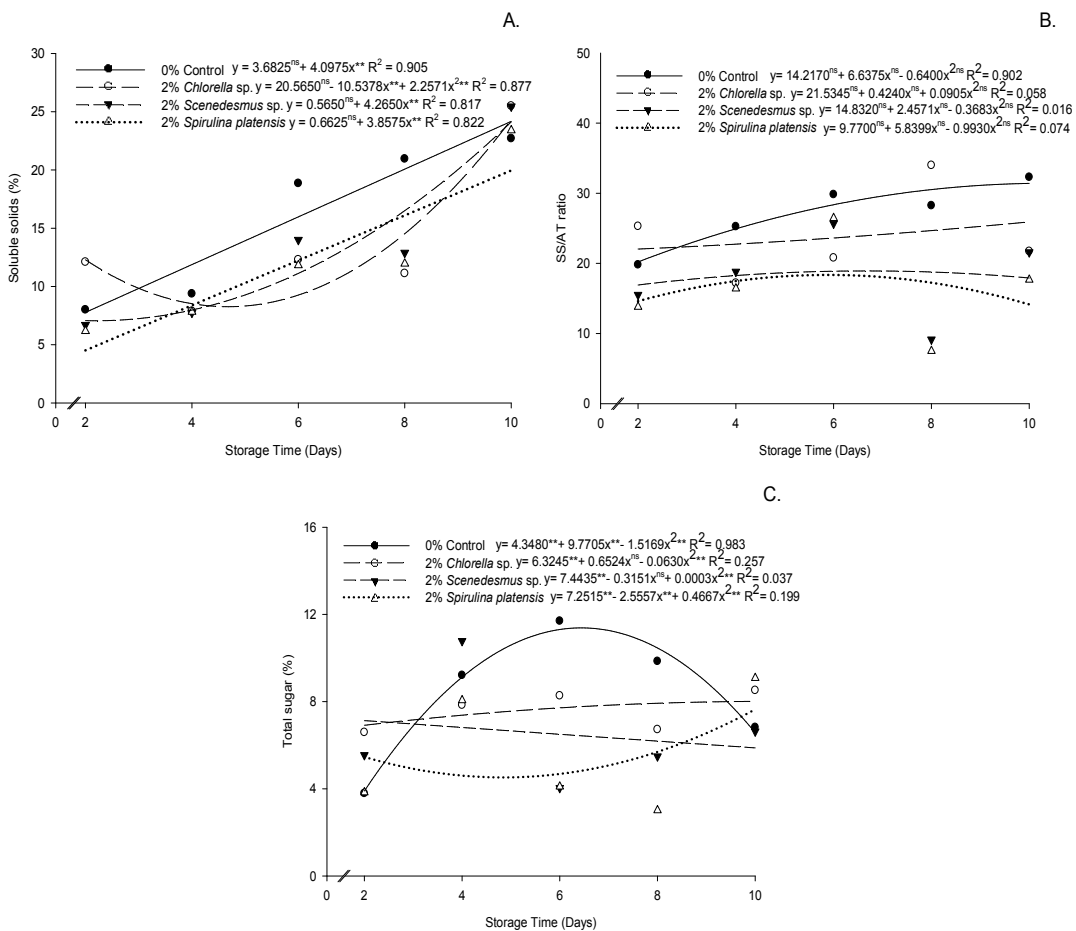


Fig. 5. Soluble solids (%) (A), SS/TA ratio (B), and total sugar (%) (C), in 'Prata Anã' bananas submitted to different coatings with microalgae base during the 10 days of storage at $25 \pm 2^\circ\text{C}$ and $65 \pm 5\%$ RH

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.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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