

Development and application on strawberries of edible coatings based on yam and corn starch added with Rio Grande cherry

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Abstract

This work aimed to evaluate the properties of biofilms based on corn and yam starch incorporated with Rio Grande cherry (Eugenia involucrata DC) and its encapsulates and use them as a coating on strawberries. The extraction of phenolic compounds from the Rio Grande cherry was optimized, and total flavonoids, antioxidant, and antimicrobial activity were analyzed for the best point. Five compositions of the film-forming solution based on yam and corn starch were evaluated and analyzed for thickness, opacity, moisture content, solubility, antioxidant activity and antimicrobial activity. In addition, antifungal analyses, water loss and color change were performed on the strawberries with and without coating. The cherry extract had a total phenolic content of 526.85 mg EAG 100 g⁻¹ and high antioxidant activity (16.99 µM Trolox g⁻¹ in ABTS; 31.71 mM ferrous sulfate g^{-1} in FRAP; 94.96% in β -carotene assay), as well as inhibition of *Escherichia coli*, *Staphylococcus aureus* and Klebsiella pneumoniae microorganisms. Adding 2% of starch and 20% of plasticizer in relation to the starch had the best overall results. Films with 0.5% fruit content had greater antioxidant activity than those with 0.03% encapsulate. Film thickness increased when encapsulated agents or fruit were incorporated, but opacity decreased. The solubility of the films changed from 0.34% for corn starch to 0.30% for encapsulate and 0.37% for fruit; yam starch films were completely soluble. Strawberries with yam starch film had the lowest water loss. Films without plasticizers showed colony formation on the seventh day of analysis. Thus, it is suggested that incorporating Rio Grande cherry fruit and its encapsulates in edible toppings could be feasible.

Keywords: Shelf-life, bioactive compounds, antioxidant activity, coating, complex coacervation, biofilms.

Graphical Abstract



Yam- and corn-based starch coatings can be applied to enhance the shelf life of strawberries

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1. Introduction

Edible films emerged from the appeal to reduce the generation of materials that are difficult to degrade and from the search for ecologically viable alternatives. Its application in food is recognized as a conservation method, as it can increase the shelf life of fruits and vegetables, maintaining fresh appearance, firmness and brightness, and increasing commercial value. In addition, active packaging with antimicrobial activity has been considered promising in reducing deterioration (Cheng et al., 2019; Durango-Villadiego et al., 2005; Farias et al., 2012; Vicentini et al., 1999).

Specific agents are used in the formulation of films, including polysaccharides, lipids and proteins. Films based on polysaccharides present excellent barriers to lipids. Those produced using starch stand out, as they are renewable natural resources. In addition to the low cost, they are odorless, tasteless, colorless, non-toxic, and show high mechanical resistance (Falcão et al., 2022; Narvaez-Gómez et al., 2021; Vicentini et al., 1999).

Starch is a reserve carbohydrate from plants. Its composition depends on the botanical origin, and it is formed by two polysaccharides, amylose (linear) and amylopectin (branched). The proportion between amylose and amylopectin influences the characteristics of the starch and, consequently, its applications. Yam starch has been used for some purposes, including the production of films, as it has a high amylose content and interesting technological characteristics, such as stability at high temperatures under low pH values (Cheng et al., 2019; Mali et al., 2004; Narvaez-Gómez et al., 2021). In addition, corn starch, when heated with water, forms a gel with good consistency, helping to prepare various foods that need to increase their viscosity. Its high amylose content simplifies its use in films (Corradini et al., 2007; Falcão et al., 2022; Weber et al., 2009; Whitt et al., 2002).

Falcão et al. (2022), Kumar et al. (2021) and Rajapaksha and Shimizu (2021) point out the importance of incorporating antioxidant and antimicrobial materials in the formulation of starchbased films, which act to ensure conservation and improve their properties. Among the various fruits that can be used to enhance the characteristics of edible films, no studies have been found with the incorporation of the Rio Grande cherry (Eugenia involucrata). The Rio Grande cherry (cereja-do-riogrande in Portuguese) is a fruit from Southern Brazil, a promising raw material for research due to its natural antioxidant compounds. Furthermore, Infante et al. (2016) and Nicácio et al. (2017) showed that both its leaves and fruit are good sources of phenolic compounds.

Given the above, the present work aimed to develop and characterize edible films based on commercial corn starch and yam starch incorporated with bioactive compounds from Rio Grande Cherry for application in strawberries.

2. Material and Methods

2.1 Materials

Corn starch and strawberry were obtained from the local market in Jandaia do Sul, Paraná, Brazil, in 2021. Rio Grande cherry was harvested in the municipality of Carambeí, Paraná, Brazil, during September and October 2020. All reagents used were analytical grade.

2.1.1 Obtaining yam starch

The yam starch was extracted according to the methodology proposed by Daiuto et al. (2005), with modifications. The yam peels were previously sanitized in an active chlorine solution at 100 ppm and were chopped and ground in a ratio of 2 kg to 3 L of cold distilled water and subsequently sieved (Tyler 100 mesh) twice. The permeate was refrigerated at 8 °C for decantation for 24 h, centrifuged (SL-700, Solab, Brazil) for 10 min at 6000 rpm and dried in a BOD incubator (LUCA-162/01, Lucadema, Brazil) at 35 °C for 24 h.

2.2 Methods

This work was divided into three parts. The first step was the extraction, characterization and encapsulation of bioactive compounds from the Rio Grande cherry; the second was the development and characterization of edible coatings based on corn starch and yam; and the third, the incorporation of Rio Grande cherry and bioactive compounds encapsulated in edible coatings and application in strawberries.

2.2.1 Optimization of the extraction of bioactive compounds and characterization of the Rio Grande cherry extract

Rio Grande cherry was sanitized in a 100ppm active chlorine solution, with subsequent rinsing under running water. Then, the seeds were removed, frozen, lyophilized (Liotop L101, Brazil) at 100 μ m Hg and –50 °C, and ground in a ball mill (SP-38, Splabor, Brazil) to prepare the extract. The extraction of phenolic compounds from the Rio Grande cherry was evaluated using a 2² central composite rotational design under the conditions shown in **Table 1**. For this, 1 g of dry sample and 50 mL of the extracting solution were added and left in an orbital shaker (TE-4200, Tecnal, Brazil), at room temperature, at different times. After this procedure, the samples were centrifuged (SL-700, Solab, Brazil) at 5000 rpm for 5 min, filtered and analyzed. The best extraction condition was determined based on the concentration of phenolic compounds, and this condition was used for the work sequence.

 Table 1 Experimental design to optimize the extraction of bioactive compounds from Rio Grande cherry

Factors			Levels		
Factors	-1.41	-1	0	1	1.41
Ethanol concentration (% v/v)	0.5	15	50	85	99.5
Time (min)	13	40	105	170	197

The content of phenolic compounds was determined the Folin-Ciocalteau by spectrophotometric method (EEQ9011I.UV-B, Drawell, Brazil), using gallic acid as a reference standard, as described by Swain and Hills (1959) and adapted by Sousa et al. (2011). In addition, the optimal point was used for the analysis of total flavonoids using quercetin as a standard (Woisky and Salatino, 1998), antioxidant activity by the ABTS *+ method (Rufino et al., 2007), oxidation of β -carotene by linoleic acid (Rufino et al., 2006a) and iron reduction (FRAP) (Rufino et al., 2006b), and antimicrobial activity using disk diffusion methodology (Ortrosky, 2008). All analyzes were performed in triplicate.

2.2.1.1 Encapsulation by complex coacervation of compounds extracted from Rio Grande cherry

The encapsulation of bioactive compounds extracted from the Rio Grande cherry was performed according to Nori et al. (2011) with some modifications. First, aqueous solutions were prepared in the proportions of 5 g 100 mL⁻¹ for gelatin (GE) and 5 g 100 mL⁻¹ for gum arabic (GA), which contained Rio Grande cherry extract (0.02 g mL⁻¹). Then, the polymeric pair was composed of GE and GA in a ratio of 1:1.

Initially, the gelatin solution was dispersed in a Tecnal shaker (model TE-102, Brazil) at approximately 10,700 rpm for 2 min. Next, GA solution was added in the respective proportion, stirring for another 2 min. While the system was stirring, the pH was adjusted to 3.5 with 1N hydrochloric acid. The system was then cooled and left to decant overnight for phase separation. Finally, the supernatant was removed, and the particles were placed in Petri dishes, frozen and lyophilized at -50° C and 100 µm Hg.

2.2.2 Development and characterization of the edible coating with and without Rio Grande cherry

2.2.2.1 Development of the filmogenic solution

Films based on yam starch and corn starch were prepared using five different methodologies, aiming at using the best result to proceed with the work. A part of the film-forming solution was dispersed in plastic Petri dishes and dried in an oven with forced air circulation (LUCA-82/480, Lucadema, Brazil) at 35 °C for 24 h for further analysis, and another part was used to cover strawberries. For each methodology, 3 samples were separated.

The first methodology (M1) was described by Fakhouri et al. (2012) and performed with some modifications. First, a proportion of 3 g starch/100 mL of distilled water, with 10% sorbitol in relation to the mass of starch, was used. This solution was heated at 85 °C in a water bath for approximately 5 min to the starch gelatinization. Subsequently, it was allowed to cool at room temperature (25 °C), and the strawberries were immersed for 1 min. Finally, it was removed and left separately to dry on a bench with natural ventilation.

The second (M2), third (M3) and fourth (M4) methodologies were described by Santos et al. (2011), with some modifications. They consist of heating water to 70 °C and placing the starch under constant agitation, using 3 proportions (1, 2 and 3 g starch/100 mL). Subsequently, the fruits were immersed for 1 min and set aside to dry on a bench with natural ventilation.

The fifth methodology (M5) was adapted from Santos et al. (2011) with Fakhouri et al. (2012). It uses 100 mL of heated water with 2 g of starch to gelatinize the starch, plus 0.4 g of sorbitol.

For incorporation, crushed Rio Grande cherry and the coating with the best result of the five methods tested were homogenized in a magnetic stirrer until the visual appearance was no longer altered. For the encapsulate, homogenization was performed with a glass rod so properties would not be released immediately after mixing. The films were developed with a content of 0.5% of fruit or with 0.03% encapsulate.

2.2.2.2 Characterization of the film

After drying, the part of the filmogenic solution dispersed in Petri dishes was removed and submitted to the following analyses: thickness, opacity, moisture content, water solubility, antioxidant activity and antimicrobial activity.

Thickness was measured using a digital caliper (Western Professional Line, Brazil). Fifteen measurements were performed for each film.

Opacity analysis was performed according to Kalaycioğlu et al. (2017). First, the film was cut to 3×0.99 cm to fit at the end of the cuvette, and the empty spaces did not interfere with the result. Then, a UV-visible spectrophotometer (EEQ9-111.UV-B, Drawell, Brazil) with a wavelength of 600 nm was used. The result was expressed in mm⁻¹.

Moisture content was evaluated by drying a piece of film measuring 1×1 cm in an oven at 80 °C for 24 h until constant mass. Initial and final weights were measured.

For water solubility, the film was divided into pieces of approximately 2 cm², and one of these was separated for analysis. Subsequently, this piece was immersed in 50 mL of distilled water and shaken for 24 h in an orbital shaker (TE-4200, Tecnal, Brazil) at a temperature of 25 °C at 190 rpm. Finally, the film was dried in an oven for 2 h at 80 °C. The mass was measured at the beginning and end of the experiment.

The antioxidant capacity was evaluated using the methodology of β - carotene/linoleic acid (Rufino et al., 2006a). The antimicrobial capacity was the same used for the characterization of the Rio Grande cherry described by Ortrosky (2008).

2.2.2.3 Shelf life of strawberries

The strawberries were sanitized in a 100ppm active chlorine solution, with subsequent rinsing under running water. The water present on the surface of the strawberry was removed at room temperature. The coating was applied by immersing the strawberry for 1 min and drying it at a controlled temperature (25 °C) until the end of the experiment. Strawberries with filmogenic solutions were analyzed according to weight loss, pH, titratable acidity, appearance, change in color and antifungal analysis.

Weight loss was evaluated using coated strawberries (starch-based, encapsulated and with

fruit) and without coating at 25 °C. The experiment lasted 7 days, being weighed every day on a semianalytical scale to calculate the weight loss, according to Fakhouri et al. (2012), with some modifications.

The pH of strawberries immersed with the edible coating was measured after product maceration and dilution in distilled water on day 0 and after 7 days of follow-up.

The titratable acidity was analyzed according to the methodology of IAL (2008), using 10% fruit in relation to distilled water with 3 drops of 1% phenolphthalein and titrated with a standardized 0.1 N NaOH solution. The result was given as mg of citric acid per 100 g⁻¹ pulp.

The soluble solids were measured according to IAL (2008), where the fruit pulp was homogenized with water, and the soluble solids (in °Brix) were analyzed in a refractometer. The analysis was carried out with the fruit *in natura* and after 7 days with the coated and uncoated fruit.

The appearance was visually evaluated for 7 days, checking whether or not there were signs of deterioration during this period, such as discoloration on the surface of the food, appearance of liquids and unpleasant odor.

The methodology described by Assis and Leoni (2003) for the antifungal analysis was followed, which indicates the visual verification of colony formation during storage.

2.3 Statistical analysis

The results were submitted to analysis of variance (ANOVA) at a 5% significance level using Tukey's test for analysis of means with the aid of the Statistica software, version 10. The R software (v.4.2.2) was used to perform the response surface analysis.

3 Results and Discussion

3.1 Characterization of the Rio Grande cherry

Fig. 1 shows the response surface of the optimization of the extraction of phenolic compounds from Rio Grande cherry. The best condition was found when using 180 min and 85% ethanol, which resulted in 269.19 mg EAG 100 g⁻¹. This value was close to that found by Della Antônia (2020), which was 232.43 mg EAG 100 g⁻¹. Flavonoids are among the phenolic compounds in the Rio Grande cherry, which have antioxidant activity. **Table 2** presents the

flavonoid content and the antioxidant activity of the Rio Grande cherry determined by different methodologies.



Fig. 1 Response surface of extraction optimization of total phenolic compounds from Rio Grande cherry

The flavonoid content found in the present study was higher than that reported by Della Antônia (2020) (44.94 mg 100 g⁻¹) but lower than that found by Camlofski (2008) (289.76 mg 100 g⁻¹ to 377.89 mg 100 g⁻¹). These differences are due to several factors, such as methodological procedures, climatic and geographic factors, maturation stages, and soil. However, it is expected that the Rio Grande cherry has a considerable content of flavonoids since they are a subcategory of phenolic compounds widely reported in fruits.

Table 2 Flavonoid content and total antioxidant activity by
different methods of Rio Grande cherry fruit on a wet basis.

Determination	Result				
Flavonoid content (mg 100 g ⁻¹)	84.84 ± 4.11				
Total antioxidant activity					
FRAP (µM ferrous sulfate g ⁻¹)	31,716.23±885.33				
ABTS (µM of Trolox g⁻¹)	16.99±2.61				
β-Carotene ª (%)	100.00±0.00				
β-Carotene ^b (%)	100.00±0.00				
β-Carotene ^c (%)	94.96±4.69				
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 β -Carotene assay dilutions: ^a 0.20 g mL⁻¹; ^b 0.16 g mL⁻¹; ^c 0.08 g mL⁻¹.

The Rio Grande cherry also presented antioxidant activity measured by three different methodologies. Other authors, such as Girardelo et al. (2020), Nicácio et al. (2017) and Camlofski (2008), also evaluated the antioxidant activity of the Rio Grande cherry and found high values by various methodologies, which are not directly comparable to those used in the present work.

The antioxidant activity presented by the Rio Grande cherry indicates that its use for incorporation into edible films may be feasible, causing this activity to bring about later oxidation of the material that was coated and, consequently, increase its shelf life. In addition, other applications can be indicated due to this characteristic, such as the treatment of pathologies (Cipriani et al., 2022).

Rio Grande cherry extract also shows antimicrobial activity against the different microorganisms tested, *S. aureus*, *E. coli* and *K. pneumonia*, by the halo inhibition methodology using a concentration of 0.04 g mL⁻¹ of fruit. However, on average, the inhibition generated small halos of 0.5 cm in diameter, not showing strong antimicrobial activity.

3.2 Optimization of the filmogenic solution

For the films made in Petri dishes, analyses of average thickness, opacity, solubility, and moisture content were performed, as shown in **Table 3**. It is observed that the average thickness remained between 0.04 and 0.10 mm. Hoffmann and Siguel (2018) presented in their study that higher thicknesses may be due to poor homogenization. Mali et al. (2004) and Mali et al. (2005) prepared yam starch-based films with yam starch and glycerol with thicknesses between 0.07 and 0.11 mm, similar to those in the present study.

For films based on yam starch, only that performed by M5 was not fully soluble, while the others showed total solubility in water. As for the corn starch-based films, only M4 was fully soluble, with the highest value found for M2 ($0.74 \pm 0.8\%$). As the work deals with edible coatings, the high solubility of the films is desirable. In these films, the presence of plasticizer may have contributed to the decrease the solubility. The moisture content of the films was maintained between 5 and 16%. Lower moisture content was observed in M2 and M5 for yam starch and M3 and M5 for corn starch.

Opacity is desirable in many packages when used commercially, as it retains the entry of light; however, for edible coatings, opacity may be undesirable, as the opaque edible coating retains the brightness of the fruit, making it less attractive. For corn starch, the films containing plasticizers (M1 and M5) presented the lowest opacity, a desirable characteristic in the case of edible coatings, as proposed in the present study. For formulations based on yam starch, in addition to methodologies without plasticizer, M2, with lower starch content, also had low opacity.

	Yam			
M1	M2	М3	M4	M5
0.05 ± 0.02^{b}	0.04 ± 0.02^{b}	0.05 ± 0.01^{b}	0.09 ± 0.04^{a}	0.10 ± 0.01^{a}
6.32 ± 0.91°	5.61 ± 0.49°	23.87 ± 1.87 ^a	21.80 ± 1.70 ^a	10.78 ± 1.37 ^b
11.15 ± 0.51 ^b	5.26 ± 0.20°	8.38 ± 1.45 ^b	16.36 ± 0.06^{a}	5.32 ± 1.50 ^{bc}
*	*	*	*	1.91 ± 0.01 ^a
•	Corn			
M1	M2	М3	M4	M5
0.10 ± 0.05^{a}	0.10 ± 0.04^{a}	0.05 ± 0.01 ^b	0.04 ± 0.01 ^b	0.10 ± 0.03^{a}
10.78 ± 0.01 ^d	22.75 ± 0.09°	23.69 ± 0.04 ^b	32.21 ± 0.07^{a}	9.56 ± 0.10 ^e
13.14 ± 0.00^{a}	13.21 ± 0.01 ^a	6.67 ± 0.41°	10.74 ± 0.01 ^b	6.68 ± 0.03 ^c
$0.21 \pm 0.06^{\circ}$	0.74 ± 0.08^{a}	0.41 ± 0.01^{b}	*	0.34 ± 0.03^{b}
	0.05 ± 0.02^{b} 6.32 ± 0.91^{c} 11.15 ± 0.51^{b} * $M1$ 0.10 ± 0.05^{a} 10.78 ± 0.01^{d} 13.14 ± 0.00^{a}	M1 M2 0.05 ± 0.02^b 0.04 ± 0.02^b 6.32 ± 0.91^c 5.61 ± 0.49^c 11.15 ± 0.51^b 5.26 ± 0.20^c * * M1 M2 0.10 ± 0.05^a 0.10 ± 0.04^a 10.78 ± 0.01^d 22.75 ± 0.09^c 13.14 ± 0.00^a 13.21 ± 0.01^a	M1 M2 M3 0.05 ± 0.02^b 0.04 ± 0.02^b 0.05 ± 0.01^b 6.32 ± 0.91^c 5.61 ± 0.49^c 23.87 ± 1.87^a 11.15 ± 0.51^b 5.26 ± 0.20^c 8.38 ± 1.45^b * * * Corn M1 M2 M3 0.10 ± 0.05^a 0.10 ± 0.04^a 0.05 ± 0.01^b 10.78 ± 0.01^d 22.75 ± 0.09^c 23.69 ± 0.04^b 13.14 ± 0.00^a 13.21 ± 0.01^a 6.67 ± 0.41^c	$\begin{array}{c c c c c c c } \hline M1 & M2 & M3 & M4 \\ \hline 0.05 \pm 0.02^b & 0.04 \pm 0.02^b & 0.05 \pm 0.01^b & 0.09 \pm 0.04^a \\ \hline 6.32 \pm 0.91^c & 5.61 \pm 0.49^c & 23.87 \pm 1.87^a & 21.80 \pm 1.70^a \\ \hline 11.15 \pm 0.51^b & 5.26 \pm 0.20^c & 8.38 \pm 1.45^b & 16.36 \pm 0.06^a \\ & & & & & & & & & & & & & & & & & & $

 Table 3 Physical characterization of films based on yam starch and commercial corn starch prepared using different methodologies (M1-M5)

*Totally Soluble. Different letters on the same row indicate a significant difference using Tukey's test at the 95% level.

When the filmogenic solution was applied to the strawberries, weight loss, appearance and antifungal capacity were analyzed. The mass loss was statistically the same for all methodologies, and after 7 days, there was a loss of more than 45% water for strawberries with yam-based coating and above 55% for strawberries with corn-based coating. This value also did not differ statistically for strawberries without coating. Among the strawberries coated with the 5 solutions, both for yam and corn starch, the appearance of the strawberries coated with M5 was the best after 7 days of analysis, with slight variation in color and appearance when compared to the fresh strawberry at the beginning of the study.

For the antifungal analysis, the appearance of microorganism colonies was observed on the seventh day of storage, as shown in **Fig. 2**. These colonies were visible in the strawberries coated with corn starch based on the M2, M3 and M4 methodologies. As for films based on yam starch, no colonies were formed throughout the analysis.



Fig. 2 Colony formation on the 7th day of storage of strawberries coated with (a) 1%, (b) 2% and (c) 3% corn starch with no plasticizer.

After evaluating both the film results and when applied to the strawberries, M5 was defined to prepare the coating containing the Rio Grande cherry fruit and its encapsulate.

3.3 Application of Rio Grande cherry in films

Fig. 3 shows the films incorporated with free agents and the encapsulates from the Rio Grande cherry. It can be seen that the visual characteristics of the films based on corn starch or yam starch are different. This is because yam starch is darker than corn starch due to the rapid oxidation that occurs in yam. Additionally, the films incorporated with the fruit presented the same

appearance since the fruit color interfered with the film color.

Table 4 physical presents the characteristics of films containing Rio Grande cherry. The films based on yam starch were completely soluble, either incorporated by fruit or encapsulate. However, the corn starch-based films in none of the analyzes showed 100% solubility. Solubility is an important feature when the developed film is in contact with food. Therefore, the more soluble, the more suitable the use of the film for food applications. As the objective of this study was the development of edible coatings, the film that best adapts would be that based on yam starch.



Fig. 3 Films prepared with yam starch or corn starch containing Rio Grande cherry fruit or its encapsulate. MF, film based on corn starch incorporated with fruit; ME, film based on corn starch incorporated with the encapsulate; IF, film based on yam starch incorporated with fruit; IE, film based on yam starch incorporated with the encapsulate.

However, the corn starch-based film obtained a good solubility result even though it did not show a fully soluble characteristic. Statistically, the corn starch-based films showed equal solubility, both with the incorporation of fruit and encapsulate.

Table 4 Physical analysis and antioxidant activity of films

 based on corn starch and yam starch incorporated with the

 Rio Grande cherry fruit and its encapsulate

	ME	MF	IE	IF
Thickness (mm)	0.16±0.05 ^b	0.17±0.08 ^{ab}	0.22±0.03ª	0.14±0.0 ^b
Opacity (mm ⁻¹)	1.81±0.01 ^b	1.75±0.01°	1.37±0.01 ^d	2.81±0.0 ^a
Solubility (%)	0.30±0.05ª	0.37±0.03ª	*	*
Antioxidant Activity β- carotene (%)**	12.43±4.09°	63.48±9.50ª	41.59±5.09 ^b	53.69±3.71 ^{ab}

*Totally Soluble.**At a concentration of 0.25 g film mL⁻¹. Note: Values with equal letters on the same row indicate no significant difference by Tukey's test at the 95% level. ME, films based on corn starch with encapsulate; MF, films based on corn starch with fruit; IE, films based on encapsulated yam starch; IF, films based on yam starch with fruit.

The film with the highest opacity was incorporated with the fruit based on yam starch, followed by the film with encapsulated corn starch. As the corn starch presented a better homogeneity, it adhered better to the encapsulated material and consequently presented a lower opacity.

Thickness did not show significant variation between the films, emphasizing that there are cases in which they present greater thickness due to poor homogenization during their preparation. Adding fruit or encapsulates and plasticizers increases the thickness of the film, as presented in the study by Farias et al. (2012).

Antioxidant activity was highest for films incorporated with fruit; corn starch-based film stood up among these. Furthermore, concerning the encapsulated ones, the antioxidant capacity was higher for the film based on yam starch, whose results were close to the films incorporated with the fruit. This shows efficiency in incorporating both the fruit and the encapsulate in the films since those without fruit or encapsulate (using the same methodology) presented 0.99 \pm 0.00% of antioxidant activity by the method of β -carotene.

It was found that the mass loss in strawberries with films incorporated with fruit was lower than those without coating. Contrastingly, strawberries with films based on yam starch lost less water than those with corn starch. Strawberries without a filmogenic solution have no protection from external factors, such as mechanical, physical and microbiological, and as a result, they lose water more easily. On the other hand, the strawberries with the edible coating were protected from adversities, having higher difficulty in losing water and, consequently, a longer shelf life. Water loss in strawberries showed a significant difference only after the sixth day of analysis. The water loss caused a change in the visual appearance since the strawberries without coating had a much more pronounced wilting aspect at the end of the study.

There was no colony growth during the 14 days of analysis in none of the films incorporated with the fruit or encapsulated material. Such behavior may have occurred due to the action of both the agents and the plasticizer used since this same response was observed for films without encapsulate or free agents in M5.

When evaluating the results presented in **Table 5**, it is observed that the pH did not change over the days since the initial value found for the fresh strawberry is close to that found for strawberries with encapsulates and the Rio Grande cherry. Furthermore, this value is close to that of Marques et al. (2011), which considered the strawberry ³/₄ red and ripe.

Table 5 Physicochemical parameters for strawberries with edible coatings, without coating after 7 days and *in natura* (beginning)

	pН	TSS (%)	TA (mg citric acid 100 g ^{−1} pulp)	TSS/TA ratio
В	3.64±0.01°	1.70±0.00 ^d	1.56±0.01 ^a	0.64±0.00 ^f
IF	3.58 ± 0.00^{d}	4.30±0.00 ^b	0.77±0.00 ^e	5.56±0.01°
IE	3.72±0.02 ^b	4.10±0.01 ^b	1.01±0.00 ^b	4.06±0.01 ^d
MF	3.67±0.00°	3.50±0.00°	0.92±0.00 ^c	3.82±0.01°
ME	3.91±0.01ª	5.00±0.02 ^a	0.81±0.01 ^d	6.18±0.02 ^b
in natura	3.55±0.01 ^d	5.00±0.01 ^a	0.75±0.01 ^f	6.69±0.03 ^a

Note: B, strawberry without coating; IF, strawberry with yam starch film containing fruit; IE, strawberry with incorporated encapsulated yam starch film; MF, strawberry with corn starch film incorporated with fruit; ME, strawberry with corn starch film with encapsulate. TSS, total soluble solids; TA, titratable acidity. Values with equal letters in the same column indicate that there is no significant difference by Tukey's test at the 95% level (n = 3)

Total soluble solids (TSS) can be used as a measure of the fruit's sweetness. It was noted that soluble solids in the coated samples remained close to that found for the fresh strawberry. This may have occurred because the maturation state of the fruit was already advanced considering the day of its harvest, approximately 6 days before the beginning of the experiment. Fresh strawberries presented 5 °Brix, similar to that reported by Margues et al. (2011), which found 6.9 °Brix for ripe strawberries. However, Siqueira et al. (2009) presented a soluble solids content of 9 °Brix on day 1 and 8 °Brix on day 8 of analysis for the strawberry. This slight discrepancy may be explained because when the greenest fruit is harvested, it can take longer to mature; when it matures, it has lower soluble solids than the ready-toeat harvest, as mentioned by Fagundes and Yamanishi (2001). This same behavior was observed in the present work. This was described by Kluge et al. (2002) as a result of extended storage.

The titratable acidity (TA) ranged from 0.747 to 1.565 mg of citric acid of pulp for fresh strawberries and strawberries after 7 days without an edible coating, respectively. This result was similar to that found by Marques et al. (2011), which showed 0.867 mg of citric acid per 100 g⁻¹ of pulp for the ripe strawberry. Uncoated strawberries significantly increase acidity due to changes occurring during deterioration.

The TSS/TA ratio is mainly used to determine fruit criteria such as sweetness. Françoso et al. (2008) found 6.71 and 5.97 TSS/TA ratios for strawberries on the 1st and 8th day of the analysis, respectively. In the present work, a 6.694 ratio was found for fresh strawberries, while variations of 3.819 for MF and 6.184 for ME were observed. Such results were also close to those found in the literature.

Siqueira et al. (2009) kept the strawberries under refrigeration and found an approximately 7.11 ratio after 10 days of analysis, proving that such storage conditions maintain their properties unaffected.

4 Conclusion

This work demonstrated that the Rio Grande cherry has a high antioxidant capacity and growth inhibition of S. aureus, E. coli and K. pneumoniae. The use of plasticizers for the film's development proved to be essential for better physical characteristics. Films based on corn starch and yam starch incorporated with fruit and the encapsulated agent showed good physical attributes and obtained a high level of antioxidant activity but a low level of inhibition of the aforementioned microorganisms. Furthermore, the edible coatings maintained the pH value and the soluble solids/titratable acidity ratio almost unchanged during the evaluated period. This reinforces the feasibility of using Rio Grande cherry fruit with yam starch and corn starch as a basis for preparing edible coatings and incorporating them into strawberries to increase their shelf life.

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Authors' Contributions

L. C. B. Zuge: Conceptualization; Formal Analysis; Methodology; Resources; Software; Supervision; Writing – original draft; Writing – review & editing. L. A. Alexandre: Conceptualization; Data curation; Formal Analysis; Investigation; Methodology; Resources; Software; Writing – original draft. All authors read and approved the final manuscript.

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Conflicts of Interest

The authors declare that they have no conflict of interest.

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