

Edible Coatings Based on Apple Pectin, Cellulose Nanocrystals, and Essential Oil of Lemongrass: Improving the Quality and Shelf Life of Strawberries (*Fragaria Ananassa*)

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Abstract: In this work, nine different types of edible coating based on pectin, cellulose nanocrystals, glycerol, and essential oil of lemongrass were prepared and used to coat strawberries with a film formed directly on the surface of the coated fruit. The effects of the different edible coatings on refrigerated fruits in terms of weight loss, titratable acidity, total soluble solids, pH, and anthocyanin content was evaluated after 2 days, 4 days, 6 days, and 8 days of storage. Application of the edible coatings reduced the weight loss of the coated strawberries and the anthocyanin content. The total soluble solids content of or uncoated fruit increase more markedly than that of coated fruit. In contrast, pH was maintained for both coated and uncoated strawberries. The edible coatings were effective in minimizing of the weight loss, without worsening the physical chemistry attributes. The treatments T5 and T9 presented the best results.

Keywords: Strawberry; edible coating; cellulose nanocrystals; pectin; shelf life

1 Introduction

Strawberries (*Fragaria ananassa*) are fruits with characteristic colouring, soft texture, high moisture content (about 90-95%), and slightly acidic flavour due to the presence of citric acid. The red colouring characteristic is due to the presence of natural pigments, designated as anthocyanins, which belong to the class of flavonoids, responsible for most of the blue, violet, and red colouring of flowers and fruits, whose main use in industry, is as a dye. The main anthocyanins in strawberries and blackberries are pelargonidin-3-glucoside and cyanidin-3-glucoside. The principal biological function attributed to anthocyanins, owing to their structure, is antioxidant activity [1-3].

Strawberries are consumed mainly as fresh fruit. In addition, many other strawberry products such as juices, jellies, ice creams, and cakes, among others, are found in the food industry [4]. Postharvest, they have an extremely short shelf-life because of their high metabolic activity. They are susceptible to water loss and mechanical injuries due to their soft texture [5-6]. For this reason, refrigeration is widely used to reduce spoilage and extend the shelf-life of strawberries [7].

In the literature, there are reports on the use of procedures to minimize the occurrence of the senescence process in the fruits, extending their shelf-life and maintaining their nutritional value using, for example, atmospheric-pressure cold plasma [6], low doses of radiation [8], and the application of edible coatings made from different formulations such as gelatin and cellulose nanocrystals [9], pectin, pullulan, and chitosan [10], aloe vera and ascorbic acid [11], chitosan coatings containing lemongrass essential oils (EOs) [12] and quinoa protein-chitosan [13].

Edible coatings are commonly prepared from polysaccharides, proteins, and lipids, and applied directly on the surface of the fruits to be coated, in a thin layer, which is pre-formed or formed directly on the surface of the product as a protective cover [5]. These materials act as barriers that produce a modified atmosphere, minimize respiration rate, reduce moisture exchange, delay deterioration, control microbial growth, and carry functional ingredients such as antioxidants or antimicrobials [14, 10].

The most frequently used coating process comprises dipping the fruit in a filmogenic solution for a short period of time and a subsequent drying step for film formation on the surface of the coated fruit. Thus, the formation of an extremely thin film, imperceptible to the naked eye, occurs on the surface of the coated fruit. The ingredients used in the preparation of coatings must be non-toxic, and suitable for ingestion together with the coated fruit [15].

The combination of both, edible coatings and refrigeration can increase the postharvest shelf life of strawberries; however it is desirable to minimize the weight loss, and extend the shelf life, without changing the organoleptic characteristics of the fruit [13].

Cellulose nanocrystals (CNCs) are needle-shaped particles with nanometric dimensions that can be extracted using acid hydrolysis [16-17]. The use of this procedure to isolate CNCs is based on the quicker hydrolysis kinetics presented by the amorphous regions, as compared to the crystalline ones. Thus, this process breaks the disordered and amorphous parts of the cellulose, releasing single and well-defined crystals. The main characteristics stimulating research on CNCs are biodegradability, biocompatibility, low density, and high crystallinity [17].

Pectin or pectic substances are collective names for a group of closely associated polysaccharides present in plant cell walls, where they contribute to complex physiological processes like cell growth and cell differentiation and so determine the integrity and rigidity of plant tissue. Thus, the pectin can be defined as a hetero-polysaccharide predominantly containing galacturonic acid residues, in which varying proportions of the acidic groups are present as methoxyl esters, while a certain quantity of neutral sugars might be present as side chains [18-19]. Pectins are widely used in the food industry due to their capacity to modify the viscosity of food, beverages, jams, jellies and fruit juices [20].

EOs are organic volatile constituents responsible for the fragrance of many plants. These compounds have shown great importance in certain research because they are potentially useful to minimize or control the negative effects and damage that microorganisms cause in the food industry and agriculture [21]. Nevertheless, their incorporation in food systems is mainly limited by flavour considerations, since effective antimicrobial doses may exceed organoleptic acceptance levels [22].

In this context, many antimicrobial compounds present in essential oils have demonstrated their importance for microbial population control by targeting foodborne microorganisms, so they help in the production of food products of higher quality and safety [23]. For instance, essential oil of lemongrass (EO) has antimicrobial activity against a diverse range of microorganisms including moulds, yeasts, and gram-positive and gram-negative bacteria [24,14].

However, as far as we know, there are no studies on the application of essential oil of lemongrass, cellulose nanocrystals, apple pectin, and glycerol as edible coatings for strawberries. Thus, the goal of this study was to evaluate the effects of different edible coatings on the physical-chemical parameters (weight loss, total soluble solids (TSS), pH, titratable acidity, and anthocyanin content) of strawberries in order to extend the shelf-life of this fruits.

2 Experimental Part

2.1 Materials and Methods

The Bleached kraft wood pulp of *Eucalyptus urograndis* (hybrid of *Eucalyptus urophila* and *Eucalyptus grandis*) was provided by the Conpacel Company (Limeira, São Paulo, Brazil). Pectin from apple ($M_w = 30.000-100.000 \text{ g mol}^{-1}$, degree of esterification $\geq 70-75\%$, Sigma-Aldrich), glycerol (Synth), ethanol (Synth), hydrochloric acid (Vetec), and sodium hydroxide (Vetec). The strawberries (*Fragaria ananassa*) were purchased at a local market and were selected to be uniform in size, ripening stage (three-

quarters of surface red colour), and free of visible defects, essential oil of lemongrass (Mundo dos óleos Essenciais-Brazil).

2.2 Isolation of Cellulose Nanocrystals (CNC)

The CNCs were isolated from bleached kraft wood pulp and characterized in our previous work [25]. The CNCs were prepared by hydrolysis at 45°C for 50 minutes using 20 mL of H₂SO₄ (9.17 M) per gram of fibre.

2.3 Preparation Of Apple Pectin Solutions

The suspension of apple pectin was prepared at a concentration of 1% w/v, from apple pectin powder with vigorous stirring for 48 hours.

2.4 Preparation of Edible Coatings

The formulations used were divided into two groups, the first, without glycerol, and the second with glycerol incorporate. The control (T1) refers to strawberries on to which no edible coating was applied. Tab. 1 shows the formulations, the volumes of each of the ingredients, and the number of immersion used in the application of edible coating.

Table 1: Formulations used in the preparation of filmogenic solutions for the application of edible coating onto strawberries

Name of treatment	Volume of CNC suspension (mL)	Volume of apple pectin suspension (mL)	Volume of glycerol (mL)	Volume of lemongrass essential oil (mL)	Number of immersions
T1	-	-	-	-	-
T2	18.20	230.00	-	-	1
T3	18.20	230.00	-	0.12	1
T4	18.20	230.00	-	-	2
T5	18.20	230.00	-	0.12	2
T6	18.20	230.00	0.50	-	1
T7	18.20	230.00	0.50	0.12	1
T8	18.20	230.00	0.50	-	2
T9	18.20	230.00	0.50	0.12	2

The volumes, shown in Tab. 1 were considered, taking into account that the maximum volume for the filmogenic solution should be 250.00 mL. To prepare the filmogenic solutions used in edible coatings, 7.30% v/v of CNC suspension, 0.05% v/v to OE, 0.20% v/v of glycerol were incorporated and the volume of the pectin suspension made up the balance.

The preparation of the edible coatings is quite simple. In formulations in which there was no addition of the lemongrass EO, the volumes of suspensions of pectin and CNC; CNC and glycerol were submitted to magnetic stirring for 1 hours. For formulations with added EO, the EO was mixed with glycerol, then with the suspension of pectin, and finally with CNC suspension. This mixture was subjected to vigorous magnetic stirring for 2 hours.

2.4.1 Application of Edible Coatings

Approximately 500 ripe strawberries of uniform size and free of physical damage and fungal infection were used. Strawberries were dipped in water dried at room temperature and stored for later application of edible coating, which was done by dipping the fruits for two min, in the respective formulations shown in Tab. 1 for a duration of two min. After the set time, the strawberries were placed in a clothesline, for the drying step. This time was chosen because it is a reasonable time for formation of the coating on the surface of the fruit and because the same immersion time is given by data already published in the literature [7].

For formulations where the dipping processes were performed twice, a first layer of the coating was applied and the fruits were then subjected to the process of dipping again for two min followed by a subsequent drying step. Afterwards, they were dried at room temperature under natural convection for 3-4 hours and then cold-stored in polystyrene packaging and coated with poly-vinyl chloride film. The packaging with strawberries was stored in a refrigerator at $10^{\circ}\text{C} \pm 1^{\circ}\text{C}$. This procedure was performed for the characterizations that will be presented.

2.4.2 Extraction of Juice

For juice extraction, 500 g of each sample (coated and uncoated) was ground in a blender followed by filtration to remove solid contents. Juice from the fruit was used to determine the total soluble solids, pH, titratable acidity, and anthocyanin contents.

2.5 Weight Loss (%)

The strawberries (coated and uncoated) were stored in a refrigerator at $10^{\circ}\text{C} \pm 1^{\circ}\text{C}$. Then they were removed from the refrigerator every two days and placed in an environment with controlled temperature ($23^{\circ}\text{C} \pm 1^{\circ}\text{C}$) in order to evaluate the weight loss according to our previous work [25]. After weighing, the strawberries were put back in the refrigerator. This assay was performed in triplicate and the visual appearance and weight loss of the samples were assessed during eight days.

2.6 Puncture Test Measurement

Strawberry samples were cut longitudinally and analysed using a Texture Analyser (TA XT plus, Stable Micro Systems). A cylindrical probe 5 mm in diameter was moved perpendicularly at a distance of 5 mm from the strawberry surface at a constant speed of 1 mm s^{-1} until it passed through the strawberry. Force-displacement curves were recorded until rupture of the film and were used to determine puncture strength (N) caused by the addition of edible coatings to the surfaces of the strawberries.

2.7 Total Soluble Solids (TSS), Titratable Acidity (TA) and pH

TSS, expressed as degrees Brix ($^{\circ}\text{Brix}$), was measured with an Abbe WYA refractometer (Biocotek) calibrated against sucrose at 20°C . Titratable acidity (TA) was determined using 5.0 mL aliquots of extract of the fruit in 2.5 mL of distilled water and titrated with 0.1 mol L^{-1} NaOH to an end-point of pH 8.1. The monitoring of the pH change was done by pH meter (pH 2221 Hanna Instruments) and a magnetic stirrer was used for mixing. TA was expressed as grams of citric acid per 100 g of juice. The pH value was obtained through direct measurement of the juice by direct immersion of the electrode of a pH meter (pH 2221 Hanna Instruments). This assay was performed in triplicate.

2.8 Anthocyanin Contents

The anthocyanin content of strawberries was determined using a spectrophotometric method. An aliquot of 1 mL of juice was diluted into 12 mL of the solvent solution of 95% ethanol and 1.5 N HCl, in a ratio of 85: 15, [5], then this mixture was stored for approximately 30 minutes, and covered with aluminum foil. The anthocyanin content was calculated from the absorbance at 510 nm (UV 250 1 PC spectrophotometer).

The anthocyanin concentration was estimated as pelargonidin-3-glucoside, using a molar absorptivity coefficient of $36.000 \text{ L cm}^{-1} \text{ mol}^{-1}$, and was expressed as milligrams of pelargonidin-3-glucoside per 100 g of fruit in accordance with [26]. This assay was performed in triplicate.

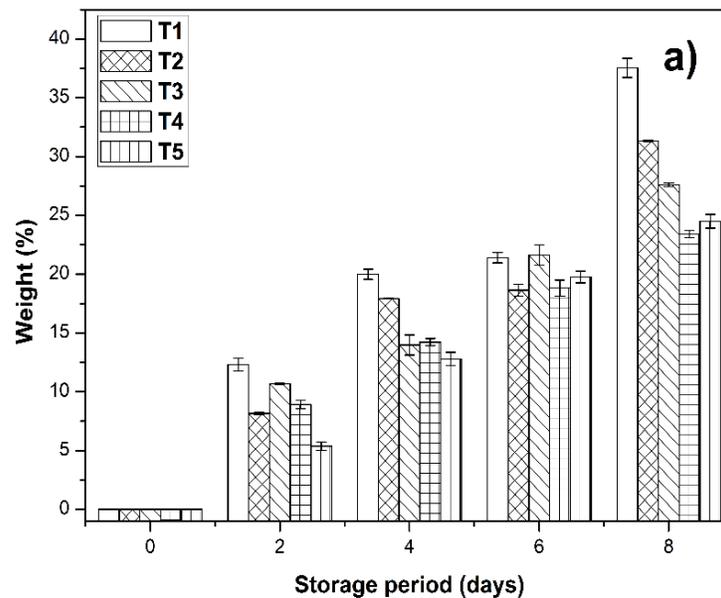
2.9 Statistical Analyses

Statistical analysis was carried out with the software (R 3.3.2 System Statistical Analysis). The test was conducted using a completely randomized design with a factorial arrangement 9×5 , where 9 refers to treatments used for edible coating of strawberries and 5 refers to the number of days of storage, that is, 0 days, 2 days, 4 days, 6 days, and 8 days. The variables evaluated were weight loss, anthocyanin content, pH, soluble solids, and titratable acidity. To verify the normality of the data and homogeneity of variances, the Kolmogorov-Smirnov and Bartlett tests were used, respectively. Then the analysis of variance and comparison of means were carried out using the Scott-Knott test for the variables: weight loss, anthocyanin content, and TA, at a 5% probability level. Because the variables: pH and TSS did not pass the normality assumption, a non-parametric analysis of variance was performed using the Kruskal-Wallis test followed by Dunn's test.

3 Results and Discussion

3.1 Weight Loss

Graphics of the weight loss for strawberries coated without glycerol (Fig. 1(a)) and with glycerol in the formulations (Fig. 1(b)) show that all fruits exhibited a progressive weight loss during storage. Normally, weight loss occurs during fruit storage due to the respiratory process, the transfer of humidity and some oxidation processes [27].



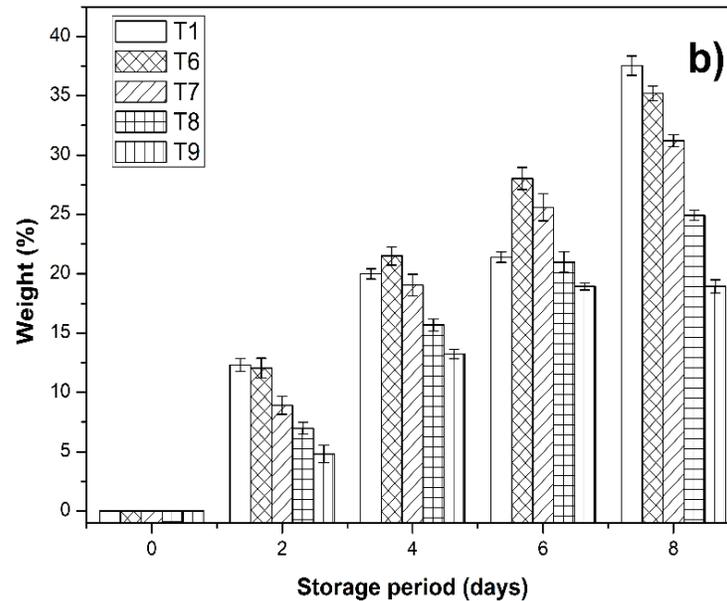


Figure 1: Graphic of weight loss strawberries coated with the formulations used in the preparation of edible coatings (a) without glycerol addition; (b) with glycerol addition

The weight loss obtained for the control (T1) was higher than that of the coated strawberries, on every day of storage, independently of the applied coating. Controlling the mass loss of fruit through the application of edible coating is a major goal of coating technology. In this way, the film formed on the surface of strawberries retards the migration of moisture from the fruit to the environment, thus reducing weight loss during storage. The adding of lemongrass EO to the coating formulations as a lipid source could be responsible for the lower weight loss of coated samples. Similar data were obtained by Azarakhsh and colleagues using lemongrass EO incorporated into alginate-based edible coating for fresh-cut pineapple [28].

The formulations, for which the dipping process was performed twice, (T4, T5, T8 and T9) resulted in a smaller weight loss of the coated strawberries. These results are in accordance with literature [29], who investigated edible coatings based on wheat gluten for refrigerated strawberries. Fakhouri and colleagues observed the maximum loss around 35% in strawberries coated with CNCs and gelatin [9]. In this manner, on the sixth storage day, all treatments evaluated in this study, led to smaller values of weight loss, as revealed by the data displayed in Figs. 1(a) and 1(b).

In this sense, when comparing all treatments over time, some features' mean values were quite close to one another and so the statistical analysis of results was realized for a better discussion of the results obtained in terms of the analysed parameters. The results (Tab. 2) show that depending of the treatment used, the weight loss values are statistically different ($p < 0.05$).

On the second day of storage, the treatments T1, T3 and T6 were not effective in minimizing weight loss. On the fourth day of the storage, the same trend was observed: treatments, T1 and T6 led to a smaller reduction in terms of weight loss. On the last days of storage, the treatment T6 showed the lowest effectiveness, followed by T1, in minimizing the loss of mass of the coated fruits. The differentiation can be made on the basis of the different composition of the treatments studied (T1 and T6).

Among all the treatments evaluated, the edible coatings that presented the best values in the minimization of the weight loss were T2, T4, T7, T8 and T9. Our results were consistent with studies previously reported in the literature, showing a reduction in the weight loss for coated fruit. The coating acts as a protective semipermeable barrier that slows the exchange of gases between the fruit and the environment, reducing respiration, water loss, and oxidation reactions and thus contributing to prolongation of the shelf life of the fruit [30,10].

Table 2: Effect of edible coatings by treatments on weight loss, Titratable acidity, pH, anthocyanin content and total soluble solids

Treatments	Days of storage	Weight loss ¹ (%)	Titratable acidity ¹ (g citric acid/100 g)	pH ²	Anthocyanin contents ¹ (mg/g)	Total Soluble Solids ² (°Brix)
T1	0	0	0.95 ^a	3.39 ^a	8.73 ^c	6.08 ^a
	2	12.31 ^a	0.90 ^a	3.48 ^a	11.98 ^a	7.08 ^a
	4	19.99 ^a	0.75 ^b	3.59 ^a	16.96 ^a	8.33 ^a
	6	21.39 ^c	0.64 ^c	3.74 ^a	18.30 ^a	9.58 ^a
	8	37.54 ^a	0.45 ^c	3.89 ^a	18.53 ^a	11.33 ^a
T2	0	0	0.88 ^b	3.50 ^a	6.82 ^d	6.33 ^a
	2	8.14 ^b	0.84 ^b	3.47 ^a	7.97 ^c	6.58 ^b
	4	17.92 ^b	0.81 ^a	3.45 ^a	8.87 ^d	7.08 ^b
	6	18.63 ^d	0.75 ^b	3.43 ^a	9.56 ^e	7.08 ^b
	8	31.31 ^c	0.66 ^a	3.46 ^a	11.47 ^e	8.67 ^b
T3	0	0	0.93 ^a	3.50 ^a	8.33 ^c	6.33 ^a
	2	10.68 ^a	0.87 ^a	3.52 ^a	9.56 ^b	6.67 ^b
	4	13.97 ^d	0.83 ^a	3.56 ^a	11.30 ^c	7.08 ^b
	6	21.62 ^c	0.81 ^a	3.63 ^a	12.49 ^c	7.33 ^b
	8	27.60 ^d	0.69 ^a	3.63 ^a	13.04 ^d	9.08 ^c
T4	0	0	0.87 ^b	3.54 ^a	6.32 ^d	6.42
	2	8.91 ^b	0.85 ^b	3.57	8.07 ^c	6.58
	4	14.23 ^d	0.79 ^a	3.59	8.95 ^d	6.58
	6	18.82 ^d	0.66 ^c	3.62	10.99 ^d	7.33
	8	23.41 ^e	0.60 ^b	3.66	14.45 ^c	8.33
T5	0	0	0.84 ^b	3.54 ^a	6.83 ^d	6.00 ^a
	2	5.37 ^d	0.74 ^b	3.49 ^a	6.96 ^d	6.58 ^b
	4	12.78 ^d	0.72 ^b	3.48 ^a	7.92 ^d	7.08 ^b
	6	19.75 ^d	0.69 ^c	3.46 ^a	8.94 ^c	7.58 ^b
	8	24.48 ^e	0.66 ^a	3.44 ^a	11.09 ^e	8.83 ^b
T6	0	0	0.94 ^a	3.49 ^a	11.59 ^a	5.58 ^b
	2	12.05 ^a	0.93 ^a	3.46 ^a	12.73 ^a	6.58 ^b
	4	21.49 ^a	0.83 ^a	3.44 ^a	14.20 ^b	6.91 ^c
	6	28.02 ^a	0.78 ^a	3.42 ^a	14.03 ^b	7.50 ^c
	8	35.19 ^b	0.63 ^a	3.45 ^a	16.35 ^b	7.80 ^d
T7	0	0	0.94 ^a	3.50 ^a	9.93 ^b	6.25 ^a
	2	8.91 ^b	0.84 ^b	3.52 ^a	11.20 ^a	6.83 ^b
	4	19.04 ^b	0.79 ^a	3.56 ^a	11.70 ^c	7.08 ^b
	6	25.59 ^b	0.72 ^b	3.62 ^a	11.36 ^d	7.32 ^c
	8	31.20 ^c	0.63 ^a	3.64 ^a	11.44 ^e	9.08 ^c
T8	0	0	0.84 ^b	3.53 ^a	8.78 ^c	6.83 ^c
	2	6.98 ^c	0.81 ^b	3.54 ^a	10.06 ^b	6.58 ^b
	4	15.68 ^c	0.78 ^a	3.59 ^a	11.01 ^c	6.42 ^c
	6	20.97 ^c	0.66 ^c	3.61 ^a	12.24 ^c	7.33 ^c
	8	24.91 ^e	0.58 ^b	3.68 ^a	12.14 ^e	8.33 ^b
T9	0	0	0.98 ^a	3.55 ^a	7.32 ^d	6.00 ^a
	2	4.80 ^d	0.83 ^b	3.49 ^a	6.82 ^d	7.08 ^a
	4	12.85 ^d	0.72 ^b	3.50 ^a	8.22 ^d	7.08 ^b
	6	18.42 ^d	0.60 ^d	3.44 ^a	9.02 ^e	7.42 ^c
	8	18.93 ^f	0.57 ^b	3.44 ^a	12.16 ^e	8.83 ^b

¹Data expressed as the mean of three data points (n = 3); values with the same letter are not significantly different at $p > 0.05$ using the Scott-Knott test. The values represented (a-d) for each time interval indicate the range from higher to lower rank.

²Data expressed as the mean of three data points (n = 3) for the same column followed by different letters were significantly different at 0.05 using the Dunn's test.

3.2 Titratable Acidity (TA) and pH

TA has been expressed as milligrams of citric acid per gram of fresh weight, and can be directly related to the amount of organic acids present in the fruit [5]. As can be seen in Tab. 2, TA decreases with storage time of strawberries, independently of the treatment used for the coating. A reduction in acidity may be expected as a result of metabolic changes in fruit or due to the use of organic acids in the respiratory process during the storage period [5,29,31].

Some treatments showed no significant differences ($p > 0.05$) with storage period. The treatments T1, T3, T6, T7, and T9 showed no significant differences at time zero ($p > 0.05$). On the second day of storage, the samples T1, T3, and T6 presented no significant differences ($p > 0.05$), to the fourth day of storage T2, T3, T4, T6, T7 and T8; T3 and T6 for sixth day and finally in last day the treatments T2, T3, T5, T6 and T7, respectively. Similar results were obtained by Peano and collaborators, who found that TA was not affected by storage and no differences were observed between treatments with starch corn film and polypropylene perforated films [32]. Valenzuela et al. obtained similar results for TA, when using edible coating of quinoa protein-chitosan for refrigerated strawberries [13].

The effect of coatings on the TA content of strawberries without and with glycerol in the coating formulations during storage is shown in Figs. 2(a) and 2(b), respectively.

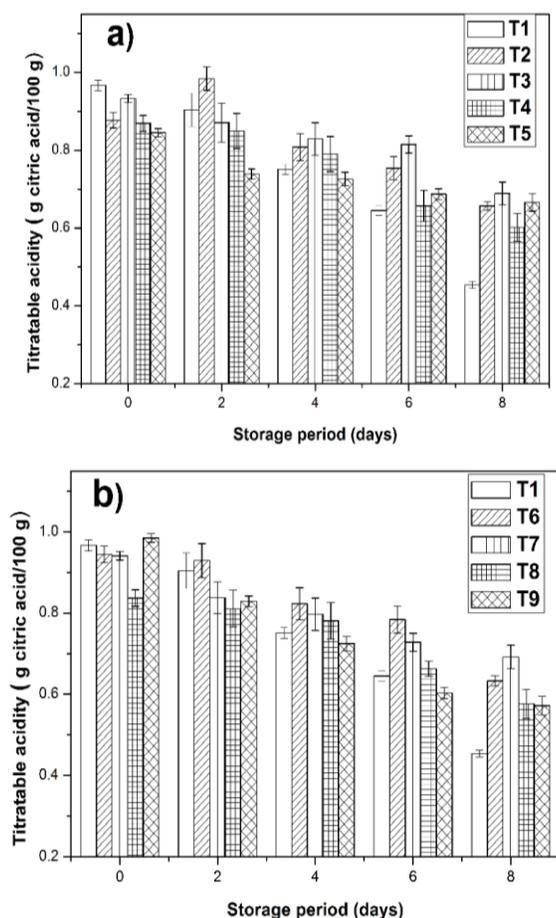


Figure 2: Graphic of titratable acidity of strawberries coated with the formulations used in the preparation of edible coatings (a) without glycerol addition; (b) with glycerol addition

Figs. 3(a) and 3(b) show the effect of different edible coatings on the pH of strawberries evaluated during the storage period.

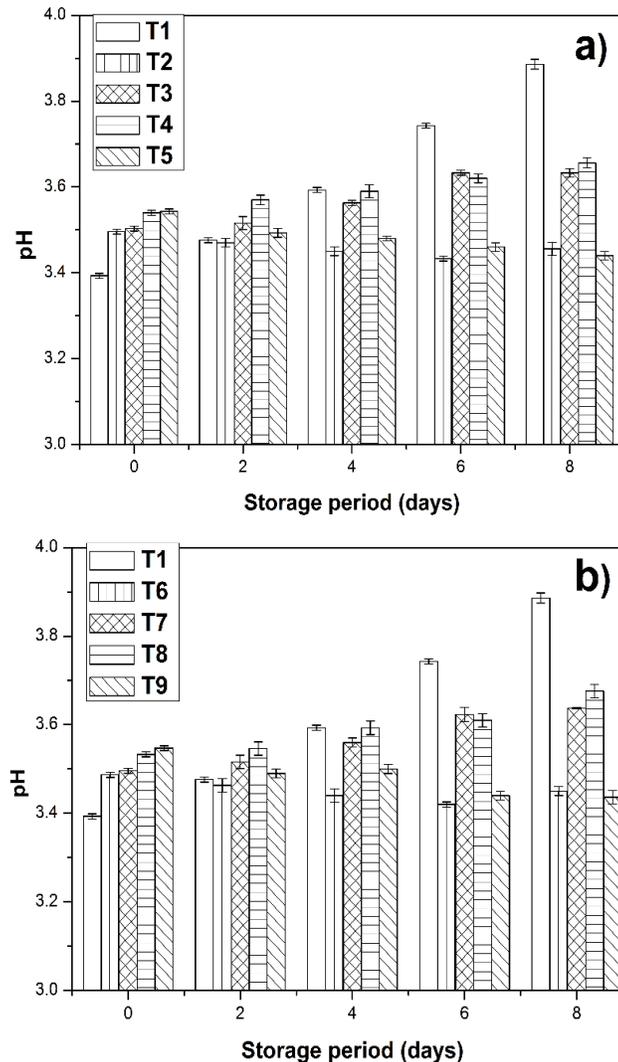


Figure 3: Graphic of pH of strawberries coated with the formulations used in the preparation of edible coatings (a) without glycerol addition (b) with glycerol addition

The values of pH of coated and uncoated samples remained constant until the last day of storage and consequently, no significant differences ($p > 0.05$) were found among the evaluated treatments, as can be seen in Tab. 2. Similar data were obtained by [10] using pectin, pullulan and chitosan as edible coating for strawberries. However, other authors have reported finding significant differences between strawberries coated with chitosan and uncoated ones [5, 33].

From the application point of view, it is quite interesting that the utilization of edible coatings on strawberries did not cause significant changes in the pH. In this sense, the TA decreased over time, as expected, but when considering a storage time of two to three days, the time on which this fruits are consumed in natura, the treatments T3 and T6 showed no significant differences from the control T1. These results indicate that the application of these coatings did not change the chemical composition of the fruit drastically or prevent its consumption.

3.3 Anthocyanin Content

Fig. 4(a) shows the UV vis spectrum for the juice obtained from the uncoated strawberry at start of the storage time at pH 1. The content of anthocyanins in uncoated and coated strawberries during storage are presented in Figs. 4(b) and 4(c).

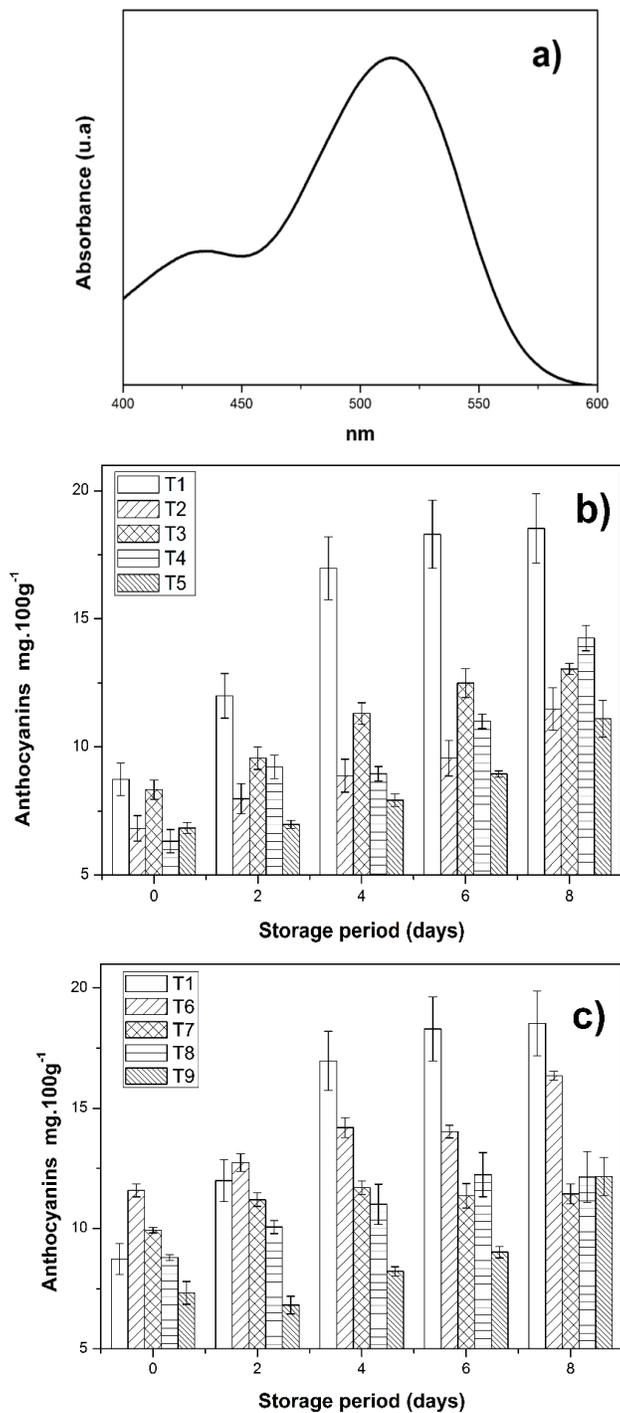


Figure 4: Representation of UV-vis spectrum (a); graphic of anthocyanins content for strawberries coated with the formulations used in the preparation of edible coatings without glycerol addition (b); (c) with glycerol addition

During the storage period, both the control and the coated fruits showed a significant increase ($p < 0.05$) in anthocyanin content, as illustrated in Tab. 2 due to synthesis of anthocyanin pigment, which contributes to the red color of the strawberry [5]. Garcia et al. reported that the greater anthocyanin content presented by uncoated samples can be explained by the higher respiration rate, especially during the last storage days. The higher the respiration rate, the more the metabolism increases, resulting in a higher pigment production [33].

The highest values of anthocyanin content found for the control (T1) can be related to weight loss greater than that which occurred in the uncoated fruit. So, a possible modification of the atmosphere in the coated samples due to application of edible coating could explain this behaviour.

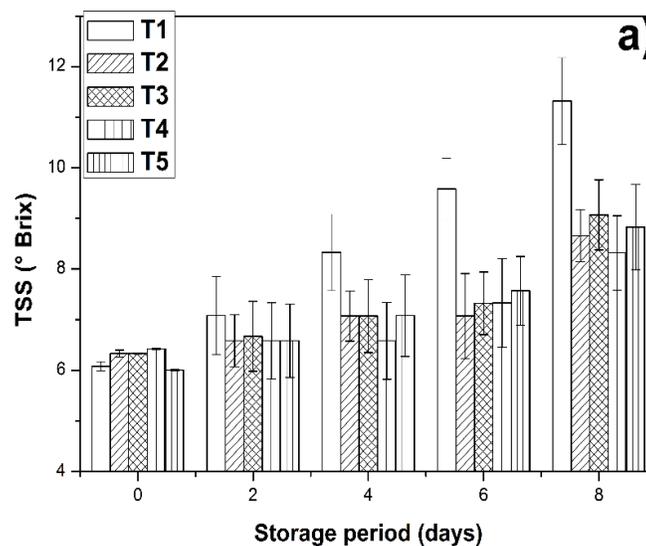
At zero days, T6 exhibited a higher anthocyanin content than expected, which may be attributed to the degree of maturation of the fruits used. When the was analysed for two days of treatments, the samples T1, T6, and T7 had higher levels of anthocyanins. In this line, with an increase in the working days, only treatment T1 brought higher values, than the other treatments used, with $p < 0.05$.

Anthocyanins are water-soluble pigments that confer a range of attractive colours on fruits, flowers and leaves. The amount of anthocyanin is important for the evaluation of the maturity of strawberries because the index of ripeness used for harvesting is the redness resulting from the anthocyanin synthesis. According to Shin et al., the anthocyanin content of strawberries increased significantly after four days of storage at 20°C; however, no changes in pigment concentration were observed when samples were stored at 0.5°C and 10°C [34]. Garcia et al. reported that uncoated samples presented a higher pigment concentration, when compared with coated samples at each storage time [33].

The modification of the colour occurs during the post-harvest ripening and the fruits become redder and darker during the storage time, due to the synthesis of anthocyanins. This is in agreement with the results obtained by [35,36] who reported that during storage, the fruits became redder and darker due to synthesis of anthocyanins.

3.4 Total Soluble Solids (TSS)

The effect of coatings on the TSS of strawberries is shown in Figs. 5(a) and 5(b) for strawberries coated without and with glycerol, respectively during the storage period. TSS is expressed as degrees Brix. The TSS parameter in strawberries is related to the degree of maturity and basic metabolic reactions that increase the sugar content and fruit sweetness throughout the storage period [10].



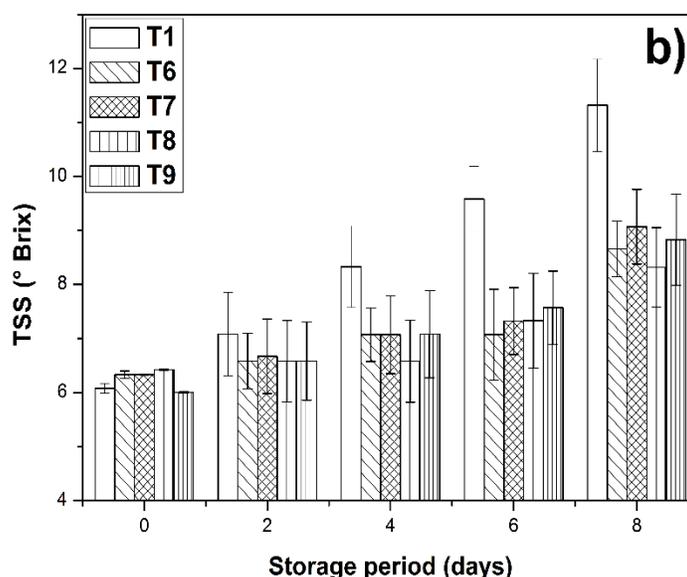


Figure 5: Graphic of Total Soluble Solids (°Brix) of strawberries coated with the formulations used in the edible coatings preparation (a) without glycerol addition (b) with glycerol addition

All samples were analysed during a storage period of the days, because the fruit is usually consumed within this time. The values of TSS differ very little from one another, indicating that the application of edible coatings the fruit surface did not cause significant changes in the values of TSS of the samples, with $p > 0.05$.

The TSS of uncoated strawberries (T1) increased progressively with the storage period. This sample presented more pronounced TSS because the sample did not provide protection against the external atmosphere. The coated samples showed the same trend as the uncoated sample, but the values differed slightly from each other when the treatments were compared directly. In this context, the application of edible coatings to the strawberry's surface can help maintain the TSS, because the protective barrier formed hampers the occurrence of gas exchange between the fruit coated and the external atmosphere, contributing to maintenance of the TSS coated fruit when compared with the control, as a function of storage time.

After the eighth storage day, the significant weight loss represented by the water loss that occurred in uncoated strawberries may have contributed to an increase of the TSS value for this sample compared with other samples. Similar data were obtained by Velickova et al. using edible chitosan-beeswax coatings on the quality of fresh strawberries [31].

TSS was analysed because it is one of the most important parameters analysed by consumers when purchasing these fruits. Thus, the application of the coatings does not interfere in the consumption of coated fruits.

3.5 Puncture Test Measurement

Tab. 3 shows the data obtained for maximum strength for the puncture of strawberries given by (N).

The results showed that all of the coatings applied to the strawberries caused changes in the force required to puncture them. Accordingly, these results confirm that an extremely thin film formed on the surface of the fruit, and acted as a protective barrier, and in general, it is necessary to apply a higher strength to puncture coated the fruits. Similar data were obtained by Amal et al. for strawberries coated with soy, or wheat gluten protein, as a carrier of thymol and calcium chloride. The coated strawberries required higher puncture force than the control sample [35].

According to the data shown in Tab. 3, when comparing the formulations in the presence and absence of glycerol and EO which the dipping process performed only once (1X) the results shown that, incorporation of glycerol and EO in the reduction of the maximum force values for puncture of the coated strawberries. When the dipping processes were performed twice (2X), the results are very close together. This behaviour may be related to the number of immersions and not to the ingredients used in the preparation of edible coatings.

The values shown in Tab. 2 were obtained for the coated strawberries at zero storage time. There are reports in the literature, regarding evaluation of the texture of this fruit, according to the maintenance of firmness with storage time, and there was a decrease in texture of strawberries. This confirms that the senescence process inevitably occurs during the storage time [10].

Table 3: Values of maximum force employed for puncture of strawberries (N) for each edible coating applied to strawberries

Name of Treatment	Force employed for puncture of strawberries (N)
T1	0.1281 ± 0.0714
T2	0.2548 ± 0.0266
T3	0.2047 ± 0.0194
T4	0.2084 ± 0.0134
T5	0.1563 ± 0.0067
T6	0.2276 ± 0.0071
T7	0.1521 ± 0.0091
T8	0.1989 ± 0.0063
T9	0.1411 ± 0.0124

4 Conclusions

It was possible to form a thin film on the surface of the coated fruit regardless of the type of formulation applied according to the results obtained from the puncture test measurements. Compared with the uncoated fruits, the coated fruits presented better behaviour with regard to weight loss over long periods. In this way, the treatments T5 and T9 minimize the mass loss. Regarding the pH values, there were no significant differences between the uncoated and coated fruits. Titratable acidity values decreased relative to storage time for both uncoated and coated fruits. The anthocyanin content increased significantly in the uncoated fruit, as consequence of the larger weight loss of this sample. In this manner, the anthocyanin content of coated fruits increased less markedly, which can be attributed to the protective barrier formed during the coating of the fruit. The total soluble solids content of or uncoated fruit increase more markedly than that of coated fruit. This can be attributed to the edible coating applied on the surface of the fruit, which helps in maintenance of the total soluble solids. Thus, the edible coatings were effective in minimizing of the weight loss, without worsening the physical chemistry attributes.

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