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Original Article

Strawberries treated with biodegradable film containing plant extracts

Morangos tratados com filme biodegradável contendo extratos de plantas

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Abstract

Strawberry (*Fragaria* x ananassa Duch.) is a highly perishable fruit whose characteristics make it susceptible to developing microorganisms. Plant extracts have been studied as an alternative to pesticides to control spoilage microorganisms, responding to the expectation of the population seeking a healthier way of life. The fungus *Botrytis cinerea* is a facultative pathogen of vegetables, which can affect all stages of the development of several fruits, such as the strawberry, where it causes gray rot. *Trichilia catigua* (catuaba), *Paullinia cupana* (guarana), *Stryphnodendron barbatiman* (barbatimão), and *Caesalpinia peltophoroides* (sibipiruna) are planted in the Brazilian flora and have demonstrated pharmacological properties in their extracts. This work aimed to treat strawberries with a biodegradable film containing extracts of these species to evaluate strawberry conservation. There were notable distinctions in mass loss between the extract-treated and control samples. The pH, total acidity (TA), and soluble solids parameters exhibited consistently significant means across both sets of samples. Luminosity increased over the course of days in the color parameters, with the exception of strawberries coated with guarana. The red color showed greater intensity, except for those coated with barbatimão extract. Considering the results, it is possible to conclude that the coatings used can become an alternative to enhance the conservation of strawberries.

Keywords: fungal deterioration, *Botrytis cinerea*, conservation.

Resumo

O morango (*Fragaria* x ananassa Duch.) é uma fruta altamente perecível cujas características o tornam suscetível ao desenvolvimento de microrganismos. Extratos vegetais têm sido estudados como alternativa aos agrotóxicos para o controle de microrganismos deteriorantes, atendendo a expectativa da população que busca um estilo de vida mais saudável. O fungo *Botrytis cinerea* é um patógeno facultativo de hortaliças, podendo afetar todas as fases do desenvolvimento de diversas frutas, como o morango, onde causa a podridão cinzenta. *Trichilia catigua* (catuaba), *Paullinia cupana* (guarana), *Stryphnodendron barbatiman* (barbatimão) e *Caesalpinia peltophoroides* (sibipiruna) são plantados na flora brasileira e têm demonstrado propriedades farmacológicas em seus extratos. Este trabalho teve como objetivo tratar morangos com filme biodegradável contendo extratos dessas espécies para avaliar a conservação do morango. Houve diferenças significativas entre as amostras de extrato e controle para perda de massa. Os parâmetros pH, acidez titulável e sólidos solúveis mantiveram a mesma média significativa entre as amostras. Houve aumento da luminosidade ao longo dos dias nos parâmetros de cor, exceto nos morangos revestidos com guaraná. A cor vermelha foi mais acentuada, exceto para os revestidos com extrato de barbatimão. Considerando os resultados, é possível concluir que os revestimentos utilizados podem se tornar uma alternativa para potencializar a conservação dos morangos.

Palavras-chave: deterioração fúngica, *Botrytis cinerea*, conservação.

1. Introduction

Small fruits, such as strawberries, have increased production due to the appreciation by consumers for their sensory attributes and nutritional value, highlighting the presence of vitamin C, folic acid, bioactive such as polyphenols and beta-carotene, in addition to minerals and other vitamins such as A, B1, and B2 (Giampieri et al., 2012). The presence of flavonoid compounds and anthocyanins in the strawberry is highlighted, whose antioxidant power can help prevent diseases, including cardiovascular and cancer (Tomic et al., 2017). The strawberry crop is

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considered prominent among the small fruits, mainly due to its wide acceptance for fresh and industrialized consumption, including sweets, yogurts, jellies, and ice creams (Contigiani et al., 2018). In Brazil, the annual production is 40 thousand tons, and the production cost for strawberries is approximately R$ 9.1 million (Brandt et al., 2022).

Due to their characteristics and high respiratory rate, strawberry becomes eminently perishable, with up to 40% post-harvest losses (Aamer et al., 2021). Non-conforming handling, transport, storage, and marketing conditions also contribute to the increase in the deterioration process by microorganisms. In this way, it is essential to use technologies that help conserve its post-harvest quality (Aamer et al., 2021).

During cold storage, strawberries exhibit a high rate of degradation by a metabolic activity where attacks by microorganisms such as *Botrytis*, *Penicillium*, *Phomopsis,* and *Rhizopus* are undeniable. *Botrytis cinerea*, a fungus that frequently causes gray mold in strawberries, stands out. It is a facultative fungus, non-pathogenic to humans, which lives saprophytically in the soil and can remain dormant for long periods in the form of sclerotia. It is disseminated by its conidia through the air or by insects (Silva, 2014). The attack by *B. cinerea* is favored by mild temperatures and high humidity, becoming severe in rainy periods. During cultivation, the infection can start in the flowers and quickly reach green or ripe fruits, spreading to stems and leaves until the plant dies. However, the process can also occur post-harvest, during storage causing fruit rot (Vanti et al., 2021).

Fungicides have been used to control diseases in strawberries, whose excessive and indiscriminate use can reduce the product's quality, resulting in health risks and mental damage. This fact causes concern among consumers about food safety, restricting the market and intensifying the search for food free of pesticide residues (Herrera-Romero et al., 2017).

Plant extracts seem promising in controlling phytopathogens because they present active compounds, including alkaloids, flavonoids, isoflavonoids, tannins, coumarins, glycosides, terpenes, phenylpropanoids, and organic acids. This wide variety of combinations has the advantage of different modes of action on microorganisms, preventing the development of resistance by the pathogen (Marinho et al., 2022). Brazil is considered significant in terms of biodiversity on the planet, having about 20% of the total number of plant species. Among them are *Trichilia catigua* (catuaba), *Paullinia cupana* (guarana), *Stryphnodendron barbatiman* (barbatimão), and *Caesalpinia peltophoroides* (sibipiruna) stand out.

Trichilia catigua, or catuaba, is a plant native to the Northeast and Central Plateau regions and extends to Pará and Maranhão (Bernardo et al., 2021; Longhini et al., 2017). The pharmacognostic characterization of *T. catigua* bark shows the presence of flavonoids, free anthracenes, condensed tannins and saponins, alkaloids, mucilages, coumarins, and essential oils, which makes its study so important (Valmorbida et al., 2008).

Paullinia cupana, or guarana, a plant native to the northern region of Brazil, has been used for therapeutic purposes, mainly as a nervous system stimulant. It has antidiarrheal and diuretic properties and protective action on DNA and may prevent carcinogenesis (Faria Neto et al., 2019). They are used in the beverage industry and the composition of various food supplements, mainly due to their high caffeine content (Marques et al., 2019).

Stryphnodendron barbatiman, or barbatimão, is a plant native to the cerrado and fields of Brazil; they are transfers and belong to the legume family. The rinds are thick, rich in tannin, phlobaphenes, and soluble carbohydrates, with an astringent effect. Condensed tannins stimulate the healing process, have vasoconstrictor and anti-inflammatory properties, stimulate the growth of the epidermis, help re-epithelialization, and have antimicrobial and anti-ulcerogenic action (Fonseca et al., 2018; Ishida et al., 2006).

Caesalpinia peltophoroides, or sibipiruna, is an extensive ornamental plant with timber potential native to Brazil. It is used in folk medicine as an antihypertensive and has shown anti-inflammatory activity of its flowers in animal models (Pereira et al., 2021).

The four plant species presented have microbial and antioxidant activity substances, such as alkaloids, flavonoids, tannins, glycosides, and terpene. In this way, they become alternatives for controlling spoilage by microorganisms in foods (Dotto et al., 2014; Konaté et al., 2015; Ritter et al., 2021). These phytochemicals can formulate biodegradable films and edible coatings, making these species promising for the food industry (Costa et al., 2019; Pinto Andrade et al., 2018; Rabêlo et al., 2014).

Fungicides such as iprodione, copper oxychloride, procymidone, and thiophanate-methyl are traditionally used to combat B. cinerea. However, the fungus has become increasingly resistant to them. Due to this, studies that provide new options for combating the fungus have been encouraged (Marín-Chacón et al., 2017).

Given these considerations, this study aimed to coat strawberries with biodegradable films containing catuaba, guarana, barbatimão, and sibipiruna extracts and to evaluate their physicochemical characteristics and conservation.

2. Material and Methods

2.1. Material

Strawberries (San Andreas cultivar), collected directly from the producer, were selected based on ripeness, color (commercial ripening stage, approximately 80% red), and size (average length of 5 cm) to ensure homogeneity. The fungus *Botrytis cinerea* CCT 1252 from the André Toselo collection was used to evaluate the minimum inhibitory concentration. Crude extracts of *T. catigua*, *P. cupana*, *P. pluviosa*, and *S. astringens* were used at a concentration of 1%, added directly to the RPMI medium.

2.2. Methods

2.2.1. Determination of the Minimum Inhibitory Concentration (MIC)

The minimum inhibitory concentration (MIC) was determined according to the NCCLS document M38 A

(NCCLS, 2002), with some modifications. To perform the MIC, 12 test wells containing the RPMI medium were used.

The crude extracts were obtained according to methodologies described by Longhini et al. (2013) for *T. catigua*; Klein et al. (2013) for *P. cupana*; Bueno et al. (2014) for *P. pluviosa*; and Ishida et al. (2006) for *S. astringent.*

The fungi were inoculated on DG18 agar for seven days at 25 °C to promote growth and spore production. Spores were extracted by scraping the colony after the addition of saline solution with 0.1% Tween 80. The spores were counted using a Neubauer chamber and diluted to a final concentration of 10^4 spores/mL. This was the inoculum used for determining the Minimum Inhibitory Concentration (MIC).

The extracts were added directly to the RPMI medium at a concentration of 1%. For each extract, 500 µL of RPMI medium was added to wells 2 to 10 and 1000 µL to the negative control wells. In wells 1 and 2, 500 µL of each extract was added to 1 $g.mL^{-1}$. From well 2, serial dilutions of the extracts were performed up to well 10, transferring 500 µL to each test well and, in the end, discarding 500 µL from well 10.

Then, 500 µL of the suspension was added to 10^4 $spores.mL^{-1}$ of *B. cinerea* in test wells (1 to 10) and positive control (11), reducing the concentration of extracts in each well by half. The plates were gently shaken to homogenize the contents and incubated in an oven at 25 °C for 24-48 h. After this period, the growth of the fungus was verified through the turbidity of the medium and confirmed in a stereoscopic microscope.

2.2.2. Preparation of films extracted from tilapia skin

The biodegradable films were prepared using a casting technique. A solution was made containing 6 g of dry gelatin obtained from the extraction of tilapia skin, 1.2 g of sorbitol, and 300 mL of distilled water. Each film added 0, 10, 30, and 50% collagen fiber powder to its composition. This solution was stirred using a mechanical stirrer (AAKER Fistam 713D) at 640 rpm for 30 min. Then 1.2 mL of glycerol was added to the solution. The solution was at 30°C with constant stirring for 10 min, poured into silicone molds, and dried for 48h at 40°C in an oven (Venancio et al., 2019). A fisherman donated the tilapia skins in Campo Mourão.

2.2.3. Choice of extract concentration

To choose the concentration of each extract to be added individually to the film, an assay was previously carried out with the strawberries, which were sprinkled with a solution of twice and three times the MIC value in cereal alcohol. Then, after natural air drying, the treated strawberries were stored at 10 °C and observed daily for signs of deterioration. Untreated strawberries were considered as controls and treated with grain alcohol alone as white. The treatment that kept the strawberries preserved for a longer time, without beginning signs of deterioration, were solutions containing three times the MIC value, being chosen for preparing solutions with extracts and adding to the films.

2.2.4. Application of extracts in strawberries by films

Five treatments were performed on strawberries: control with biodegradable film (without extracts) and films containing extracts of catuaba, guarana, sibipiruna, and barbatimão.

The strawberries were immersed for about 30 seconds in the respective solutions; the excess was removed for about 15 seconds and allowed to dry in the laminar flow chamber, previously sterilized. Then, the strawberries were stored in polyethylene terephthalate (PET) plastic boxes, wrapped in plastic wrap, and held at 10°C for nine days. Physicochemical analyzes were performed on days: 0, 3, 6, and 9 in triplicate.

2.2.5. Physical-chemical analyzes

The mass loss of the strawberries fruit was determined by weighing them on an analytical balance at the beginning of each day of analysis. To determine the pH, a pH meter was used. Total (TA) was determined according to the methodology of the Instituto Adolfo Lutz (IAL, 2008), where a mixture of 10g of the fruit pulp was mixed with 100 mL of distilled water, and titration with 0.1 N NaOH solution was carried out until the juice reached pH 8.2 with the results expressed as a percentage of citric acid. The total soluble solids (TSS) content was measured using a refractometer, and the results were presented in °Brix.

2.2.6. Statistical analysis

The Analysis of Variance test was performed for the physicochemical analyses with a significance level of 5% with the help of the Statistica 12 program. The results were analyzed according to the Tukey test, considering testing the hypotheses that all the means of the variables of the samples would be the same or at least one of the means would be different from the others. Weight loss, pH, (TA), soluble solids, and color tests were performed on strawberries covered with extracts diluted in a biodegradable film and grain alcohol. The extracts used were guarana and catuaba. Analyzes were performed at 3-day intervals and triplicate and compared with the control sample. Principal Component Analysis also evaluated the physicochemical composition data portrayed in the OriginPro 2020b program.

3. Results and discussion

3.1. Minimum inhibitory concentration (MIC)

To determine the MIC values (Leite et al., 2023), the extract's lowest concentration that prevented the inoculum's growth under test was observed. The results are shown in Table 1.

Table 1. MIC result of crude catuaba and guarana extract against *B. cinerea*.

Extract	MIC
Guarana	0.25 $g. mL^{-1}$
Barbatimão	0.125 $g.mL^{-1}$
Sibipiruna	0.0156 $g. mL^{1}$
Catuaba	0.0312 $g.mL^{-1}$

The extract that showed the lowest inhibitory concentration was sibipiruna, followed by catuaba, barbatimão, and guarana.

Studies with *P. pluviosa* extracts are related to antimalarial activity, showing efficiency against the chloroquine-resistant strain (Kayano et al., 2011). Zanin et al. (2019) verified antimicrobial activity against *Proteus vulgaris* and *Bacillus subtilis*, inhibition of other Gram-positive and Gram-negative bacteria' growth, and anti-inflammatory and analgesic activity.

Extracts from the trunk of *Trichilia catigua* allowed us to determine the existence of protective action against lethal infections and antiviral activity in pharmacological assays in vivo. The main constituents found in these extracts include alkaloids, tannins, bitter substances, aromatic oil, resin, fatty acids, phytosterols, cyclolignans, and yohimbine. Flavalignans were also successfully isolated and tested for antimicrobial activities (Pizzolatti et al., 2002).

According to Ferreira et al. (2022), tannins are the main constituents of guarana's trunk bark and barbatimão extracts. The antimicrobial properties of these compounds are known, and one of their molecular actions is to form complexes with proteins through hydrogen bonds, hydrophobic bonds, and covalent bonds. In this way, when complexing proteins, tannins end up inhibiting extracellular enzymes of microorganisms, depriving them of the substrates necessary for their development, and acting directly on microbial metabolism, inhibiting oxidative phosphorylation (Cano et al., 2020).

In barbatimão, tannins are in the majority, constituting 25 to 30%, and due to the complexation with proteins, it performs their bactericidal and fungicidal action. The barbatimão bark extract reduced the incidence of the fungus *Fusarium subglutinans* in pineapple (Garcia et al., 2017; Ishida et al., 2006).

According to Sousa et al. (2010), the medicinal effects of guarana may be a consequence of its high concentration of alkaloids, tannins, and saponins, which is corroborated by Klein et al. (2015), who assume that the polyphenols present in the extract are directly responsible for its antimicrobial action. Studies carried out by Cano et al. (2020) also demonstrate the activity of these compounds in mechanisms crucial for the survival of the microorganism, among them, inhibition of protein synthesis, cell wall degradation, and folic acid biosynthesis, in addition to others, acting irreversibly or reversibly, corresponding to the bactericidal or bacteriostatic activity, respectively. Caffeine, present in guarana, is a pharmacologically active compound in the xanthines group. Ramos et al. (2010) studied the effects of caffeine on the growth of *A. ochraeus* fungi and toxigenic species of *Aspergillus* and *Penicillium*, verifying that caffeine has a negligible impact on the initial development of fungi, and when they reached the stationary phase growth, there was a decrease in mycelial mass.

Work has been carried out to inhibit the fungus *B. cinerea*, enhancing the results found in this study. Silva (2014), using chitosan, found a MIC of 0.01 $g.mL^{-1}$ for the fungus *Botrytis cinerea*, which inhibited both mycelial growth and germination of the fungus spores. The work showed that the direct contact of the fungi with chitosan is enough to produce the weakening and swelling of the hyphae, causing morphological changes in the microbial cell wall.

Cuzzi (2013) evaluated canola extract to control *Botrytis cinerea*. For mycelial growth, a concentration of 8.3% was obtained, with an inhibition of 34.4% on the mycelial growth of the pathogen. With increasing concentrations of extracts, there was a decrease in conidia germination.

3.2. Physicochemical analyzes

The weight loss of control and film-treated strawberries is expressed in percentage and can be seen in Figure 1.

Based on the data in Figure 1, it is possible to independently verify a significant difference between the weight loss for the control, sibipiruna, guarana, catuaba, and barbatimão extracts at 0, 3, 6, and 9 days. The most significant loss happened in 9 days of the control sample and the extracts of sibipiruna, guarana, and barbatimão. The pH showed significant differences in the barbatimão extract at 0 and 3 days for the barbatimão and catuaba extracts. In all extracts and the control sample, the alcohol had a low pH value (<4.0). Only the control and barbatimão extract in soluble solids maintained the same mean with a significance level of 5% over the nine days.

Because gelatin forms a protein-rich film, Miller et al. (2021) justify that sorbitol and glycerol-based coatings are edible, biodegradable, and promote a sound barrier against gas. They also form mechanical protection, which increases post-harvest life and minimizes food spoilage (Merino et al., 2021).

Strawberries have a sensitive epidermis that provides little protection against moisture loss, contributing to high mass loss values during storage (Contigiani et al., 2021; Duarte-Molina et al., 2016).

Adding lipid material to the hydrophilic coating and extracts was expected to improve the moisture barrier properties. However, this effect was only observed in the coverage with catuaba due to the significant presence of tannins in its composition. Thus, the mass loss of coated strawberries depends on the coating type used and storage conditions.

Hernández-Muñoz et al. (2008) observed values between 14.2% and 19.6% of mass loss with chitosan coating, stored at 10°C for 7 days. Thus, the mass loss results obtained in this study indicate that the treatments were viable (Borges et al., 2013).

Silva et al. (2011), working with yellow passion fruit residue biofilms applied to apples, found that the treatments did not prevent mass loss but showed lower values than the control. Gelatin biofilms are associated with almost all extracts.

The pH for the control and the extracts of sibipiruna, guaraná, barbatimão, and catuaba at times 0, 3, 6, and 9 days showed no significant difference, except the value that differs significantly at the time of 9 days for the extracts of barbatimão and catuaba (Table 2). The film showed a low pH value in all extracts and the control sample (<4.0). The lowest pH value compared to the control was treatment with catuaba and barbatimão in nine days.

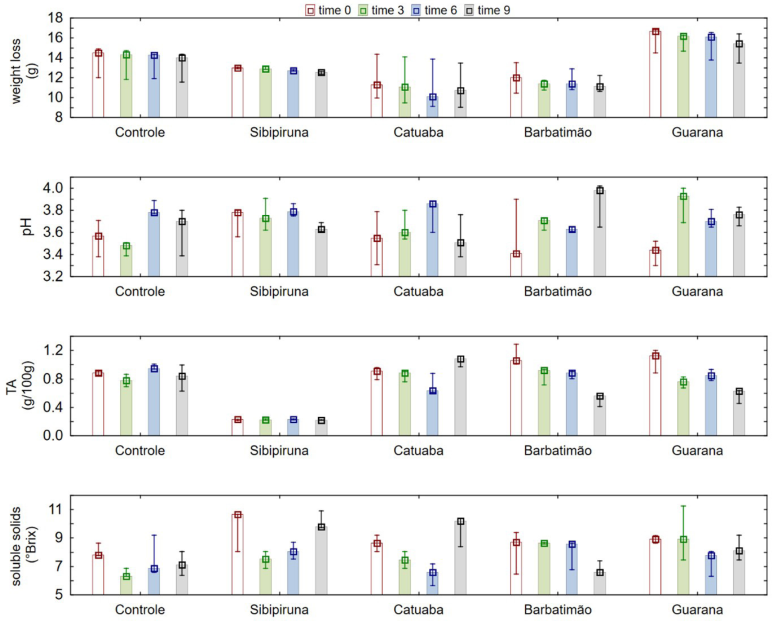

Figure 1. Physicochemical analyzes performed on covered strawberries by extracts diluted in a biodegradable film.

It is observed that after 9 days of storage, all pH values were lower than those found on the study's first day, except for the catuaba sample. The increase in pH value in samples with barbatimão extract found at nine days of storage agrees with results presented by Ponce et al. (2010), in which they reported an increase in pH in strawberries stored with PVC film.

These data are emphasized by Nour et al. (2021), who mention that the organic acids present in strawberries tend to decrease during their ripening due to their use as a respiratory substrate, which causes an increase in pH values. The treatments do not significantly alter the results because, according to the authors, organic acids are used as a substrate in respiration, altering the TA and, consequently, the pH during the softening of the fruits.

Under commercial conditions, differences in pH values can be recorded in the same cultivar, given that most producers tend to harvest even before the strawberry is fully ripe, intending that the strawberries resist until the moment of their commercialization (Costa et al., 2011).

Perdones et al. (2012) applied chitosan coating with lemon essential oil in strawberries. They observed that the pH values increased according to the maturation process, which points to the possible influence of this coating on the metabolic activity of the fruits.

Sales de Assis (2009), when studying strawberries using chitosan as a biofilm, found a marked increase in pH in the fruits on the 12th day, which can be explained by the concentration of acids due to fruit respiration. This variation demonstrates the acceleration of fruit metabolism and stability in fruits with chitosan from the 6th day of storage.

The highest value observed for TA was at time 0 for the sample of guarana and barbatimão extracts. The highest acidity values were found for the control and sibipiruna and catuaba extracts on the sixth, third, and ninth days, respectively. The lowest acidity was found for the control and extracts of barbatimão and guarana on the ninth day. Organic acids in plant tissues can be found in free or esterified forms (met hyl, propyl, hexyl, among others). In the presence of their potassium salts, free weak acids present a slight variation in pH as a function of the equilibrium established in the system. Acids are associated with potassium salts and constitute buffer systems that regulate cell enzymatic activity (Chitarra and Chitarra, 1990).

Both pH and TA are related to the determination of acids present in the food. They differ because pH measures the dissociated acid that has buffering power. At the same time, TA measures the total amount of acids present (free organic acids in the form of salts and phenolic compounds) (Dong et al., 2015; Gol et al., 2013).

The acidity content of non-climacteric fruits, such as strawberries, also tends to decrease during storage due to the respiratory process and the conversion of acids into sugars, characteristic of senescence (Chitarra and Chitarra, 1990).

Hassan et al. (2020), who studied the shelf life of strawberries using edible toppings, noticed that chitosan and chitosan toppings with calcium chloride added with oleic acid had TA, which is consistent with the present work, statistically lower than the uncovered fruits, with no difference between the covers used. Brackmann et al. (1999) assert that higher TA levels may result from a decreased respiratory rate, as acids are the substances most readily available for obtaining energy in the Krebs cycle. Moraes et al. (2008), when researching strawberries with a controlled atmosphere, found acidity values between 0.54 and 0.61 g/100g, close to those found in this study.

Table 2. Correlations between the physical-chemical parameters and the the first two principal components.

Parameters	Principal Components	
	PC1	PC2
	r	r
(a)		
mass loss	0.40476	-0.34955
pH	-0.43056	-0.24775
TA	-0.20591	0.48629
soluble solids	-0.23662	-0.51821
L*	-0.22335	0.46530
a*	0.54165	-0.04293
b*	0.45730	0.30506
(b)		
mass loss	0.31753	0.55657
pH	0.39713	-0.18220
TA	0.25997	-0.51114
soluble solids	0.09923	0.62376
L*	0.43643	0.03055
a*	-0.47751	0.07162
b*	-0.49552	0.02527
(c)		
mass loss	-0.50020	0.12623
pH	0.23240	-0.57329
TA	0.53885	-0.03173
soluble solids	0.36943	0.05946
L*	0.42260	-0.06798
a*	0.14639	0.64580
b*	0.26255	0.47874
(d)		
mass loss	-0.24549	0.48802
pH	-0.14691	0.57516
TA	-0.35105	-0.33683
soluble solids	-0.43783	-0.38933
L*	0.44444	-0.35980
a*	0.51131	0.00574
b*	0.37981	0.19105

r = Pearson's correlation coefficient.

Cells have acids that, when associated with potassium salts, form a buffer system, which enables the regulation of enzymatic activity. This system allows that, in some cases, the fruit juice enables variation in TA without variation in pH. Acidity is quickly lost when the fruits begin to ripen; however, in some cases, there may also be a slight increase in acidity values with advancing maturation (Chitarra and Chitarra, 1990).

Only the control and barbatimão extract in soluble solids maintained the same mean with a significance level of 5% over the nine days. For Jesus Filho et al. (2018), in addition to the variety, the soluble solids content is influenced by the degree of fruit maturation, being directly proportional. The increase in the range of soluble solids in the maturation process may be due to compounds' biosynthesis or polysaccharides' degradation. In their study, the authors found soluble solids values of 6.74 and 7.31°Brix. According to them, the different treatments did not favor the biosynthesis or the degradation of compounds that would affect the concentration of soluble solids.

Another important parameter is the loss of water in the fruits during storage since dehydration causes an increase in the concentration of soluble solids and TA, interfering with the increase in values during storage (Borges et al., 2013).

The organic acid content tends to decrease during maturation, due to its oxidation in the tricarboxylic acid cycle, in the respiratory process, or to its conversion into sugars since, at this stage, there is a greater energy demand due to increased metabolism (Chitarra and Chitarra, 1990).

From the data, it was observed that in all treatments applied, the values of total soluble solids (TSS) at the end of the experiment were higher at the beginning of the investigation, except for the treatment with guarana, in which an increase in the TSS content was observed in the sample. These SST variations can be attributed to the dehydration of the fruits. The significant difference presented for the variable TSS can be explained by the variation of the strawberry samples, which, despite having previously undergone a selection regarding color and size at the beginning of the experiment, showed differences in the degree of maturation between them.

The organic acids in the fruits, in balance with the sugar contents, represent an essential quality attribute. Many of these acids are volatile, thus contributing to the characteristic aroma of fruits (Françoso et al., 2008).

Analytical data of L* show that the films added to the extracts showed a slight change in the control group's lighter fruits, with more excellent brightness, making the fruit more attractive (Figure 2). This probably occurred because the formed films did not present irregularities on their surface and because of the homogeneity of the forming substances since irregularities would reduce the reflection of specular light. This result was satisfactory, as some studies showed that applying emulsified filmogenic solutions to strawberries can cause color changes and increase the opacity of the coated fruits (Vargas et al., 2006). About the parameters a* and b*, there was a slight decrease over the nine days, values justified by a change to less vivid colors, and the darkening of the shade angle of the strawberry peel is characteristic of oxidative darkening reactions that are typical of fruit senescence. The film with barbatimão extract had the lowest a* parameter value over the nine days, and the catuaba extract film had the lowest decrease in the b* parameter. A reduction in the a* parameter and an increase in b* would mean that the strawberries would turn yellow, which did not occur in the other samples. On the other hand, the higher the values of the a* parameter, the redder the strawberries, as

(Colussi et al., 2021) found when encapsulating strawberries using allyl isothiocyanate electrospun ultrafine zein fibers.

Figure 3 presents the analysis of the main components (PCA) for the physicochemical evaluation of the control samples and of the four extracts at 0, 3, 6, and 9 days. In Figure 2a, in which observations were made according to time 0, the first (PC1) and the second (PC2) dimensions explained 83.00% of the total variation. Main component

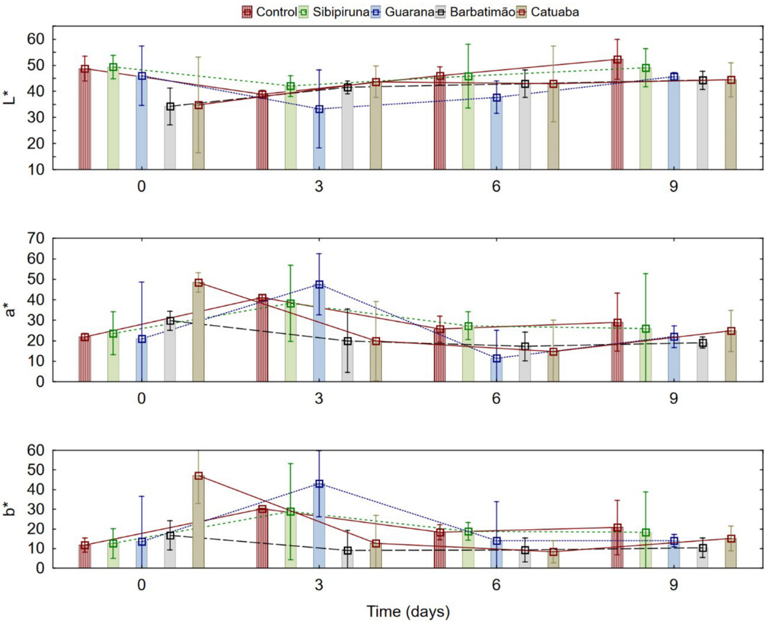

Figure 2. Color parameters for film-coated strawberries.

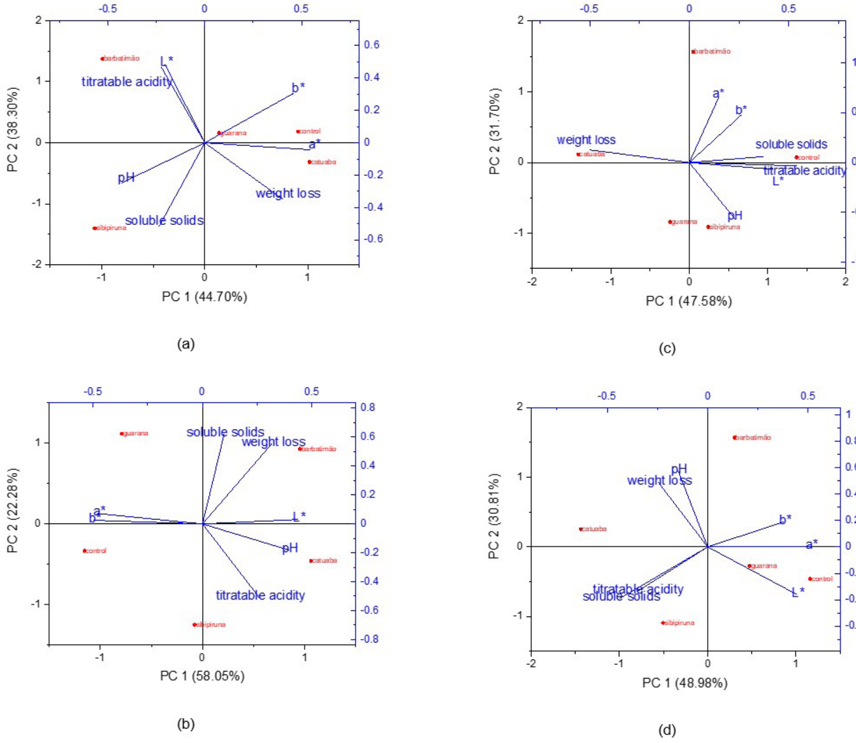

Figure 3. Principal component analysis for control samples and extracts.

1 (PC1) represented 44.70%, while main component 2 (PC2) was 38.3%. In Principal Component Analysis (PCA), variables are represented as vectors, characterizing the samples close to them. The longer the vector, the better the explanation of the variability between samples, and the values obtained by Pearson's correlation confirm the relationship between the parameters observed in the principal components analysis (Table 2a), demonstrating correlations between some variables studied at time 0. Mass loss has a negative correlation with pH (-0.43056), TA (-0.20591), soluble solids (-0.23662), and the color L* parameter (-0.22335) and positive with parameters a* (0.54165) and b* (0.45730). It is worth mentioning that at time zero, there was no mass loss for the samples.

In Figure 3b, in which observations were made for the third day, the first (PC1) and second (PC2) dimensions explained 80.33% of the total variation. Main component 1 (PC1) represented 58.05%, while main component 2 (PC2) was 22.28%. The relationship between the parameters observed in the principal components analysis (Table 2b) demonstrates correlations between some variables studied on the third day. It is worth mentioning that the mass loss began to undergo variations and presented a positive correlation between pH (0.39713), TA (0.25997), soluble solids (0.09923), and the L* color parameter (0.43643), and negative with parameters a* (-0.47751) and b* (-0.49552).

For the sixth day of treatment, it is possible to verify in Table 2c and d that there is a quadrant inversion for mass loss. While at times 0 and 3, it was in the positive quadrant of PC1, it moved to its negative quadrant at 6 and 9 days. It positively correlated with all variables involved at time six and only with the color parameters at time 9. For the times of 6 and 9 days, there was an explained variation of 79.38% (PC1 = 47.58% and PC2 = 31.70%) and 79.79% (PC1 = 48.98% and PC2 = 30.81), respectively (Figure 2c and d). For the variables pH (-0.14691), TA (-0.35105), and soluble solids (-0.43783), the correlation was negative about the mass loss on the ninth day.

4. Conclusion

Based on the results obtained, it is concluded that the crude extracts used in the tests showed inhibitory activity against the fungus *Botrytis cinerea*, with the lowest value of minimum inhibitory concentration with sibipiruna and the highest with guarana.

About time, the mass loss was lower for guarana, barbatimão, and catuaba on the third day. As for pH, the lowest values were on the ninth day for barbatimão and catuaba. The TA was lower on the sixth day for sibipiruna, barbatimão, and catuaba and on the third day for guarana. No significant differences existed between the times in each extract for soluble solids.

According to the results obtained, considering the maximum time of conservation analyzed represented by the ninth day, it is concluded that the strawberries coated with the film containing the tested extracts did not show a significant difference from the control in all physicochemical analyses. Applying these toppings added with these plant extracts on strawberries can become a viable alternative for fruit conservation during cold storage.

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