

Original Research Article

Intelligent films based on *Lobeira* fruit (*Solanum Lycocarpum L.*) starch for fresh chicken meat quality monitoring

Abstract

The use of intelligent films as colorimetric indicator devices in food packaging provides information about changes in the pH, time, temperature and microbiological properties of the product. These devices enable consumers to analyze food with the naked eye, ensuring food safety and quality. Colorimetric indicator films were produced with a polymeric matrix composed of *Lobeira* starch (AL) and the plasticizer poly(vinyl acetate) (PVA). By *casting*, indicator films containing different concentrations of vanillin (VN) were obtained from the polymeric matrix. These indicator films were characterized by FTIR spectroscopy and thermal analysis. In this study, degradation monitoring of chicken meat at 12 and 25°C was also performed. This test indicated that concentrations of 0.05% and 0.1% VN2 and VN3, respectively, provided adequate data for food monitoring.

Keywords: *Solanum lycocarpum*, Lobeira fruit, native starch, intelligent films, food quality indicators.

1. Introduction

Innovations in food packaging are rapidly developing in response to consumer demands. The packaging is characterized as an intelligent system in which information related to the quality and safety of the product can be obtained. Such systems involve the incorporation of sensors, dyes or indicator devices, which respond to changes in the initial conditions of the product by electrical, colorimetric or any other signals [1–3]. Concerns about human health in relation to deaths caused by food contamination encourage the use of indicator devices in packaging so that food can be analyzed with the naked eye by the consumer, providing greater security and an even longer life for the product [4–6]. Among the chemical compounds used as indicators in intelligent food packaging, colorimetric indicators are the compounds with the greatest potential for use in this kind of packaging. These compounds change their coloration in the presence of changes in some physical-chemical properties, such as pH [7–9], microbial growth [10–12] or temperature of the product [13–17], providing a visual indication that can be analyzed by the consumer.

Many natural extracts and compounds, as well as synthetic compounds, have been successfully used as potential colorimetric indicators [9,18–24]. Vanillin (VN), which is the main component of natural vanilla, is a natural and widely synthesized compound that has been increasingly used as a colorimetric sensor dye [25,26]. Moreover, VN is an active chemosensor against gram-positive and gram-negative bacteria, such as *Bacillus subtilis*, *Salmonella enteritidis* and *Escherichia coli*, and in the presence of these bacteria, VN coloration changes [20,27]. In addition, this material exhibits antioxidant and antimicrobial properties and therefore has great potential for use not only as a colorimetric indicator but also as a food preservative [28]. The compounds used as chemosensors for food quality indicators need to be applied in a support, mostly polymeric materials, to function properly. Thus, in recent years, biopolymers have been widely applied in the form of films to support the development of intelligent food packaging [1]. Biopolymers have the advantages of being renewable and biodegradable. Examples of biopolymers used to develop intelligent and smart packaging films are proteins [29–31] and carbohydrates [32–35].

Among carbohydrates, starch is one of the most applied in the development of intelligent, smart, and active films for food monitoring and preservation [36–40]. Therefore, native starch has great potential for application as a support to develop novel active and smart films for food packaging. As the Brazilian flora is one of the richest floras in the world [41,42], it is rich in plants that can be a source of native starches with uncommon properties [42–44]. Among these species, *Solanum lycocarpum* St. Hill (Solanaceae), commonly known as “lobeira”, is a common and abundant plant in the Brazilian Cerrado and is rich in starch [45]. This starch is well characterized in the literature because of its high content of amylose, high crystallinity, good mechanical properties, and good film-forming and morphological properties [46–49]. Thus, this starch is a promising material for biotechnological applications, including food packaging.

Hence, in this study, the viability of using lobeira starch and vanillin in the preparation of colorimetric indicator films was investigated to analyze their joint effects on the deterioration of chicken meat. Blend films made from starch and poly(vinyl alcohol) (PVA) with vanillin as a colorimetric indicator were developed. The use of these materials provides biodegradable colorimetric devices based on biopolymers, in addition to having low cost and no human toxicity, which encourages the application of these sensors in food packaging.

2. Materials and Methods

2.1 Film preparation

Films were obtained by the casting method, which consists of the dehydration of a combined starch/PVA hydrogel solution deposited on polystyrene plates with an internal diameter of 8.5 cm.

For the preparation of the films, PVA hydrogels were prepared by dissolving 1 g of polymer powder (Sigma–Aldrich Art. No. 363146, 99% hydrolysis degree) in 100 mL of distilled water under magnetic stirring at $70\pm 2^\circ\text{C}$ until complete dissolution. Lobeira starch was extracted as previously reported [49]. Then, 2 g of Lobeira starch was dissolved in 100 mL of distilled water under magnetic stirring at 60°C for 30 minutes until a hydrogel solution was formed. Afterwards, the starch and PVA hydrogels were combined at a ratio of 2:1 to obtain the final film-forming hydrogel solution. Fifty milliliters of the film-forming hydrogel solution was poured into 60 mm diameter Petri dishes to obtain a control film without vanillin.

Similarly, different concentrations of vanillin (Table 1) were added to the film-forming solution to obtain VN1, VN2, and VN3 films. The standard AL/PVA film was composed of only 50 mL of the matrix without the addition of other active compounds. Subsequently, all the films were dried in an oven at 40°C for 72 h.

Table 1. Different concentrations of AL/PVA were added to 50 mL of the polymeric matrix

Films	VN concentration (%)
VN1	0,02%
VN2	0,05%
VN3	0,1%

2.2 Film characterization

2.2.1 Infrared absorption spectroscopy (FTIR)

The FTIR spectra were obtained with a Perkin Elmer Fourier transform spectrophotometer, model Perkin Elmer Spectrometer 100, with a resolution of 4 cm^{-1} , in the region between $4000\text{--}600\text{ cm}^{-1}$, using an accessory for the total reflectance technique (ATR) with a germanium crystal (Ge).

2.2.2 Thermal analysis

Thermogravimetric analysis (TG) and differential thermal analysis (DTG) were used to determine the degradation behavior of the films. Each sample had a mass of approximately 10 mg, and TGA/DSC 1 STARe System equipment from Mettler Toledo was used. The heating rate employed was $20^\circ\text{C}/\text{min}$ with a temperature variation of $30\text{--}800^\circ\text{C}$ under an air atmosphere with a flow rate of $60\text{ mL}/\text{min}$.

2.2.3 Food monitoring test

The deterioration of the food was monitored with chicken meat, for which a plate and lid system was prepared. Five grams of chicken breast sample was placed on a polystyrene Petri dish, and under the lid, the film was fixed without contact with the meat. The films used were the VN1, VN2, and VN3 indicators, and the control film was $2\times 2\text{ cm}$ in size.

Similarly, this system was also monitored without the use of meat. The samples were placed in a DBO oven at 25°C and analyzed for five consecutive days for the first test. In the second test, the samples were subjected to 12°C for fifteen days and analyzed after 0, 1, 5, 7, 10, 12, and 15 days of storage. This test was performed in triplicate, and the changes in coloration of the indicator films were recorded by a digital camera.

3. Results and discussion

3.1 Appearance of the films

The control film and the film with VN added are illustrated in Fig. 1. Both the surface and the surface without bubbles were homogeneous (Fig. 1 (a) and (b)). Both materials have good malleability and transparency; the surface in contact with the polystyrene plate during drying presented a shiny appearance, and the surface exposed to air during drying had a matte appearance. Regarding the handling characteristics, after drying, all the films could be removed from the support plates without causing cracks or tears and could be easily handled. The different concentrations of vanillin did not affect the visual appearance of the films.

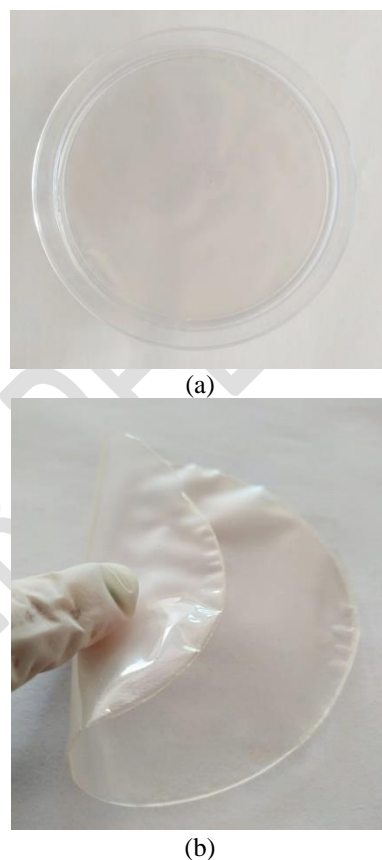


Figure 1. Appearance of the control film (a) and VN film (b)

3.2 Infrared absorption spectroscopy (FTIR)

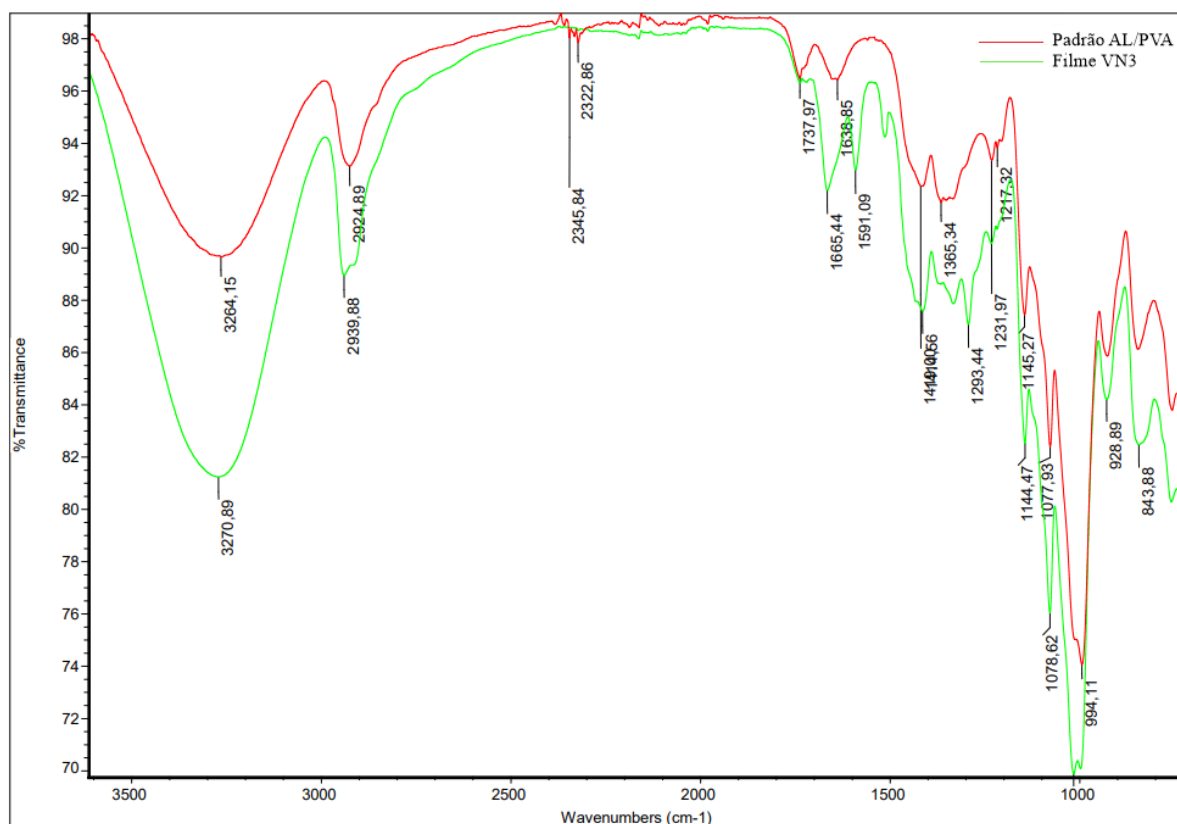
The spectra in the infrared region represent a fingerprint of a given sample forming absorption peaks that correspond to the frequencies of vibrations between the bonds of the atoms that compose the material. FTIR analyses were performed on the standard AL/PVA, MB4 and VN films.

Vanillin can be identified in a spectrum due to its characteristic molecular structure. **Fig. 2** shows the spectra of the standard film and the VN film containing vanillin. There are some differences between the spectra; a band is observed at 1665 cm^{-1} that corresponds to the

C=O stretching vibration of the vanillin aldehyde group [50], which is not observed in the standard film. The stretching vibration peaks at 1591 and 1513 cm⁻¹ in the spectrum of the VN3 film can be attributed to the aromatic ring. The peak at 3270 cm⁻¹ corresponds to the OH stretching vibration present in both spectra, and another band at 2939 cm⁻¹ is attributed to the C-H stretching vibration characteristic of the presence of [14].

The peaks at 1293 and 928 cm⁻¹ can be attributed to the bending vibration of the phenolic hydroxyl group (CHO). These peaks, except for the peak at 928 cm⁻¹, can be observed in other vanillin-containing composite films [51]. A very small shift in the absorption peak is observed, possibly due to the chemical interaction between vanillin and the polymer matrix.

Figure 2. Fourier transform infrared spectrum of the Standard AL/PVA film and the VN film of 0.1% concentration of VN in the polymer matrix.



3.3 Thermal analysis

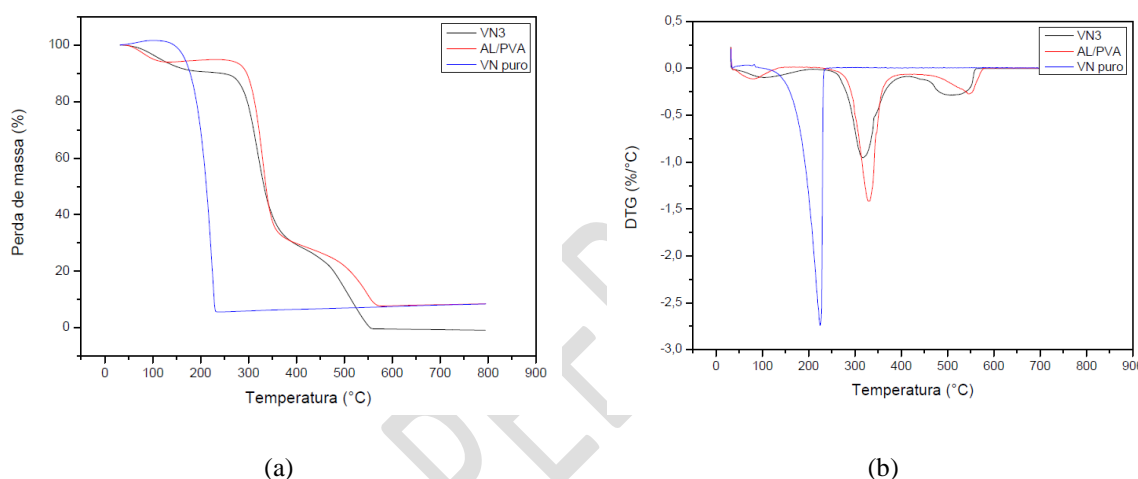
The thermal properties of the materials were investigated using thermogravimetric (TG) methods and differential thermal analysis (DTG) in the temperature range of 30-800°C under an air atmosphere.

Fig. 3 (a) and (b) present the TG and DTG curves for the pure VN materials and the control film. Three thermal decomposition steps are identified in the spectrum of the VN3 film. The first degradation step of the VN film, corresponding to the initial mass loss, occurs below 100°C, with a mass loss rate at a temperature of 109°C in the DTG curve. This step is attributed to the loss of weakly bound water, accompanied by the formation of volatile

disintegrated products, such as displaced plasticizers in the mixture and vanillin. Because pure vanillin is a volatile material, it undergoes mass loss in only one step, as shown in Fig. 6. At 130°C in the TG curve, pure VN showed a marked loss of mass corresponding to the peak in the DTG curve at 225°C. The rest of the organic portion contained in the pure VN was lost at higher temperatures, leaving no residue on the sample.

The second degradation step in the VN film starts at approximately 250°C, with the maximum mass loss rate corresponding to the peak at 318°C of the DTG curve, which indicates the degradation of starch and PVA by dehydration of the hydroxyl group. Compared to the standard film, which has a maximum mass loss rate at 330°C, the maximum mass loss rate of the VN3 film is less attenuated due to the interaction of vanillin with the polymer matrix. The third mass loss of the VN film occurs at approximately 450°C in the TG curve, with the mass loss rate corresponding to the peak of the DTG curve at a temperature of 514°C. It is assumed that this third stage of film degradation is attributed to the cleavage of the polymeric skeleton or the carbonization of the material [52,53].

Fig. 3: (a) TG curves and (b) DTG curves for the pure VN materials, control AL/PVA film and VN3 film.



3.4 Food monitoring test

The VN1, VN2 and VN3 films were subjected to microbiological monitoring of chicken meat as internal indicator devices. Internal indicators are usually placed inside the packaging and interact with the compounds present in the food. The indicators can be placed in the top space of the package or attached to the lid. For this monitoring test, a plate and lid system were prepared. Five grams of chicken breast sample was placed on a polystyrene Petri dish, and under the lid, a 2x2 cm film was fixed without contact with the meat. Similarly, this system was also monitored without using the meat.

The films used for the test were the control film and the VN1, VN2 and VN3 indicators, and the samples were named 0, 1, 2 and 3, respectively. The samples were placed in a DBO oven at the desired temperature. The test application at room temperature (25°C) was monitored for five consecutive days. **Fig. 4 (I)** shows that there was a color change in the films containing vanillin from day 3 to day 4 of the experiment. Samples 2 and 3, which contained 0.05% and 0.1% VN, respectively, showed more significant color changes on day 4 of the experiment.

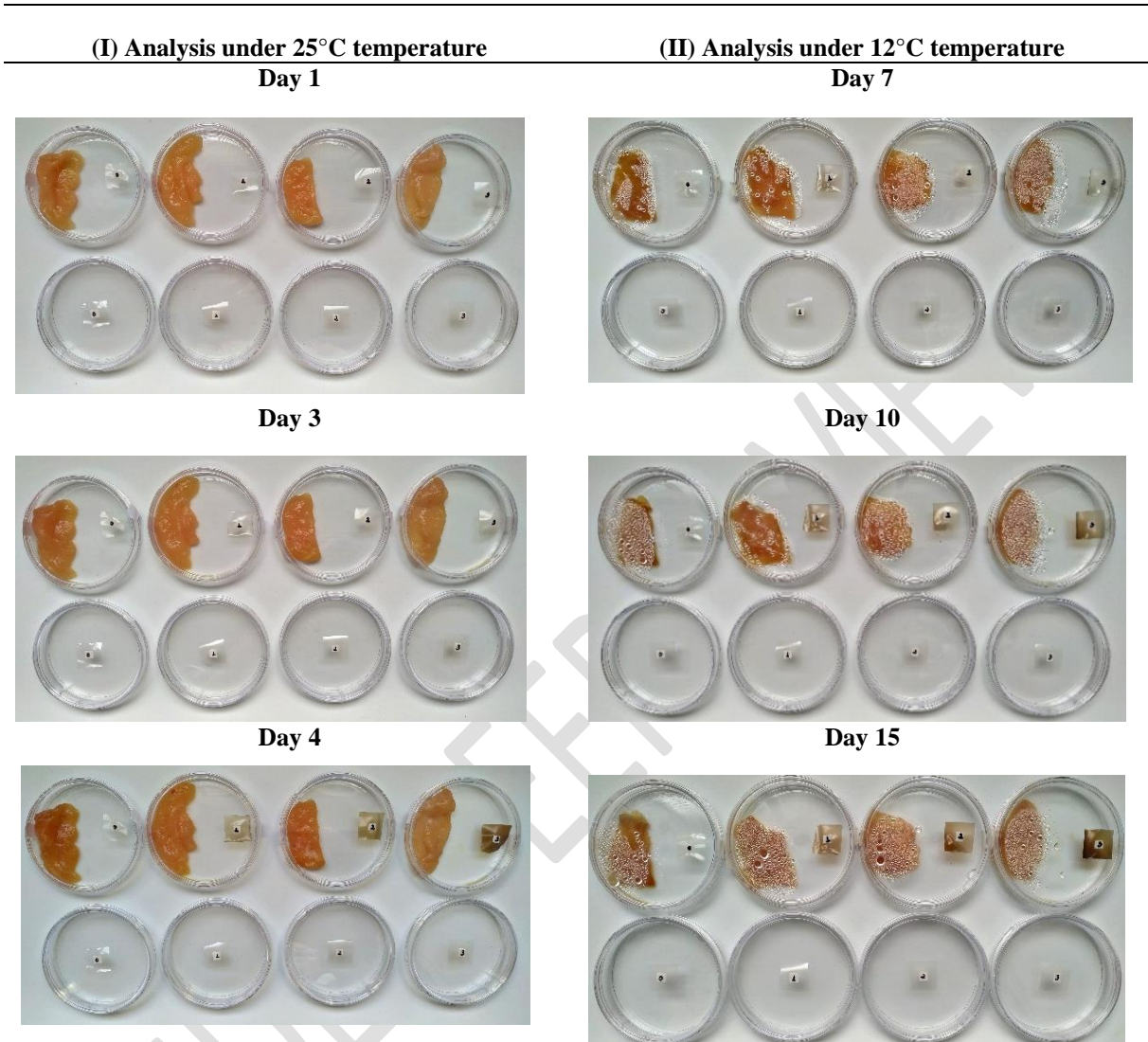
It is assumed that the greater the concentration of vanillin in the polymeric matrix of the film is, the more intense the color change due to the degradation of chicken meat. The color changes in the film indicate the probable presence of bacteria such as *Salmonella enteritidis*

and *Escherichia coli* and pathogens in chicken meat due to the accumulation of amines and ammonia, which increase the pH of the meat [54,55].

The application test was also performed at 12°C under the same conditions as those used for the previous test, and the samples were monitored during the 15-day period. Fig. 5(II) shows the test images on days 7, 10 and 15. A clear color change is observed in the VN1, VN2 and VN3 films on day 10 of the test. This color change started on day 7 of the experiment and became more intense until the last day of the test (day 15), assuming that, even at low temperature, there was proliferation and exponential growth of bacteria. These spoilage bacteria are called psychrotrophic bacteria (psychro means cold, while trophic means able to grow) because they are able to multiply in cold conditions. Fresh poultry products kept long enough at refrigerator temperatures also spoil as a result of the growth of psychrotrophic bacteria [55,56].

The films containing vanillin showed, therefore, a change in coloration against microbiological changes in chicken meat. These indicator films are independent of the pH change, but it is important to note that a fresh chicken is considered to be of very good quality at a pH of 5.8 to 6.0; when the pH is above 6.7, the meat begins to spoil and should not be consumed. In the monitoring test of chicken meat at a temperature of 25°C, the chicken meat had a pH of 7 on the third day of monitoring. In the test at 12°C, the meat pH was 7 after 8 days of monitoring. These results corroborate that there was a deterioration of chicken meat and that it should not be consumed if it has a pH of 7.

Figure 4. (I) Images from days 1, 3 and 5 of the test with chicken meat at 25°C and (II) Images from days 7, 10 and 15 of the test with chicken meat at 12°C.



4. Conclusion

The food monitoring test made it possible to observe and analyze the changes in coloration of the indicator films VN1, VN2 and VN3 in the face of the degradation of chicken meat. The indicators VN2 and VN3, with concentrations of 0.05 and 0.1% vanillin, respectively, proved to be more suitable for monitoring the deterioration of meat. The system in which the indicator was placed in the internal space attached to the lid proved to be effective for monitoring meat, suggesting that films containing vanillin are good colorimetric indicators in the presence of bacteria that cause food degradation.

Data availability

Research data are available on request.

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