

Effect of Electrolyzed Cassava Starch-Gelatin Coating on Biochemical Properties and Ripening of Banana (*Musa acuminata* L.) Fruits

Thanh Tung Pham^{1*}, Thuy Linh Nguyen², Le Phuong Lien Nguyen^{1,3}, László Baranyai¹, Khanh Son Trinh⁴

¹Institute of Food Science and Technology, Hungarian University of Agriculture and Life Sciences, Hungary

²Faculty of Fisheries, Nong Lam University, Ho Chi Minh City, Vietnam

³Institute of Biotechnology and Food Technology, Industrial University of Ho Chi Minh City, Vietnam

⁴Faculty of Chemical and Food Technology, Ho Chi Minh City University of Technology and Education, Ho Chi Minh City, Vietnam

Key words: climacteric fruit, starch, electrolysis, respiration, thin coating

In this study, cassava starch oxidized by the electrolysis was used as an edible coating to improve the shelf life of banana fruits. The effects of coating in solutions of electrolyzed starch with 1, 2 and 3% (w/v) gelatin and without gelatin addition on respiratory rate and biochemical properties of banana during 8 days of storage at room temperature (75–80% relative humidity) were evaluated. The micrographs of scanning electron microscopy showed very thin coating layers (<25.2 nm) with continuous network topology and no cracks. During the storage period, a significant reduction in respiration rate and weight loss of coated bananas compared to uncoated fruits was noted. Furthermore, the change in titratable acidity and contents of soluble solids, total carbohydrates and reducing sugars of coated bananas were slower. Increasing the gelatin content in the coating had a beneficial effect on delaying the ripening of bananas. The oxidized starch coating formulation with 3% (w/v) of gelatin demonstrated the highest efficiency as it delayed the respiratory peak 4 days more than in the uncoated bananas. This study results suggest that electrolyzed starch-gelatin coating could be a potential material to extend the shelf life of fruits.

INTRODUCTION

In recent decades, postharvest technology has been developed, in which modified atmosphere packaging and cold are widely exploited [Kudachikar *et al.*, 2011; Le Nguyen *et al.*, 2020; Radziejewska-Kubzdela *et al.*, 2007]. However, packaging made of synthetic polymers is harmful to the environment while the low temperature may cause chilling injury symptoms on fruits [Salehi, 2020; Zsom *et al.*, 2018]. Along with customer interest in biodegradable and eco-friendly materials, the edible coating is a suitable alternative to increase fruit shelf life, with the same effect as storage in a modified atmosphere [Chiumarelli & Hubinger, 2014; Karaca *et al.*, 2014; Kokoszka & Lenart, 2007].

Cassava (*Manihot esculenta* C.) starch is commonly used for fruit coating because it can be produced in large quantities at a low cost. Additionally, it is safe for human consumption and has a good gas barrier [Aguilar-Méndez *et al.*, 2008; Kim *et al.*, 2015]. However, due to the retrogradation mechanism and hydrophilicity property of starch, cassava starch-based coating becomes brittle and has a less efficient water vapor barrier [Cortés-Rodríguez *et al.*, 2020; Kim *et al.*,

2015]. Therefore, the electrolysis method is used to oxidize cassava starch to improve the physical-mechanical properties of the coating. By forming cross-linking, electrolyzed cassava starch improves the water-oxygen barrier [Trinh & Dang, 2019]. The water solubility of electrolyzed cassava starch is four times higher than that of the native cassava starch [Trinh & Dang, 2019]. Moreover, the electrolysis method also yields a higher water solubility of starch than other modification methods, which is advantageous for coating applications since it prevents coating brittleness [Basiak *et al.*, 2017; Trinh & Dang, 2019]. Furthermore, electrolyzed cassava starch is whiter in comparison with native cassava starch and the difference in color can be recognized by human eyes [Trinh & Dang, 2019]. Along with being odorless and tasteless, coating transparency is an important property that has a direct influence on consumer decisions [Agarwal, 2021]. Importantly, the electrolysis method is cheap, safe and easy to perform as chlorine can be easily washed out from cassava starch, whereas other oxidation methods may present residues of NaClO and H₂O₂ in food, or use harmful reagents such as (NH₄)₂SO₄, NaBrO₃, and KMnO₄ [Pranoto *et al.*, 2021; Trinh & Dang, 2019]. Gelatin is added to the polymer

* Corresponding Author:

E-mail: tungpt@hcmute.edu.vn (Dr. T.T. Pham)

Submitted: 22 June 2022

Accepted: 8 August 2022

Published on-line: 2 September 2022

matrix as a reinforcement to improve the physical-mechanical properties of coating [Podshivalov *et al.*, 2020]. Gelatin is a water-soluble material that is more stable in structure and less moisture-sensitive than starch. Furthermore, the addition of glycerol and sorbitol is necessary to enhance flexibility and maintain the coating integrity [Al-Hassan & Norziah, 2012; Fakhouri *et al.*, 2012]. In the starch-gelatin matrix, glycerol and sorbitol can act as plasticizers as well as compatibilizers [Podshivalov *et al.*, 2017].

Banana is a popular tropical fruit with an abundant world production [FAO, 2021]. Bananas are typically collected before fully mature and stored at room temperature. However, their shelf life is relatively short because the metabolism related to respiration and ethylene production still continues at a rapid rate after harvest [Neelam *et al.*, 2003]. There is currently a lack of studies evaluating the effectiveness of electrolyzed cassava starch coating in fruit preservation. Therefore, the purpose of this research was to develop edible coatings based on electrolyzed cassava starch, gelatin, and blends of glycerol and sorbitol as plasticizers, in order to extend the shelf life of banana during storage at $30 \pm 2^\circ\text{C}$ (75–80% relative humidity, RH). The effectiveness of electrolyzed coating was assessed by determining respiration rate, weight loss, and contents of total soluble solids, organic acids, reducing sugars, total carbohydrates, and starch.

MATERIALS AND METHODS

Materials

Banana (*Musa acuminata L.*) was purchased uniformly without any physical damage. The ripeness of bananas was selected at stage 2 based on the scale of Kader [2005]. Fruits were bought at Thu Duc wholesale market, Ho Chi Minh, Vietnam after 12–14 h harvesting.

Cassava starch powder was bought from Fococev Vietnam JointStock Company, Hochiminh, Vietnam (the purity was $\geq 98\%$, in dry basis). Gelatin (the purity was $\geq 99\%$) and glycerol (the purity was $\geq 99\%$) were purchased from Xilong Chemical Industry Incorporated Co. Ltd., Shantou, China. Sorbitol (the purity was $\geq 99\%$) was obtained from HiMedia Laboratories Pvt. Ltd. Mumbai, India.

Starch modification procedure

Starch oxidation was performed according to the method of Trinh & Dang [2019]. Cassava starch was added to 3.0% (w/v) NaCl solution to form a 10% suspension (w/v). The reaction was carried out in an electrolysis tank at room temperature for 60 min. After that, pH of the starch slurry was adjusted to 7.0 by 1 M HCl. The starch slurry was then washed by distilled water (5 times) and centrifuged ($3000 \times g$, 15 min). Finally, the starch precipitate was collected and dried in a convection dryer (50°C , 48 h).

Edible coating preparation

Before coating, banana fruits were washed with a 0.01% NaClO solution for 3 min to eliminate microorganisms on the surface and drained at room temperature (30°C) [Hossain & Iqbal, 2016]. Fruits were randomly divided into five sample groups ($n=800$). Bananas of four groups (G0–G3)

were coated with the solutions the preparation of which was described below, and the fifth group was the control (bananas without coating).

Oxidized cassava starch slurry (8.0%, w/v) was completely gelatinized at 95°C in 30 min. Gelatin was hydrated (concentrations of 5%, 10% and 15%, w/v) for 1 h at room temperature, then the gelation solutions were heated (70°C , 30 min). To obtain a gelatin-starch based coating solutions (to coat G1, G2 and G3 samples, respectively), gelatinized starch and gelatin solutions were mixed well in ratios 4:1 (v/v), thus the final concentration of gelatin in the coating solution was 1%, 2% and 3% (w/v), respectively. Parallel, coating solution starch-base only (without gelatin) was prepared to coat G0 samples. Then, plasticizers (sorbitol and glycerol) were added to all coating solutions to reach the final concentration of 1.0% (w/v) each. The coating solutions were cooled to room temperature before use.

Banana coating and storage experiment

Banana fruits were washed with water and dried at room temperature then dipped in the coating solution for 1 min. The coated and control samples were stored at $30 \pm 2^\circ\text{C}$ (75–80% RH). All samples were analyzed every day during 8 days of storage.

Microscopic analysis of coating surface

Coating layer micrographs were taken using a scanning electron microscope (SEM) to evaluate its microstructure, homogeneity, and thickness. To prepare SEM images, fruits from each treatment was randomly selected and imaged after the coating had dried. JSM 5410 LV scanning electron microscope (JEOL Ltd., Tokyo, Japan) was used at 10 kV and magnification of $1000 \times$.

Respiration rate analysis

The respiration rate of banana fruit was determined by the method of Bailey [1940] using a closed respirometer. Glass containers were assembled following Figure 1. Compressed air, with a constant flow rate (3.5 L/min), was blown throughout the system. Firstly, the air was passed through two containers (#3, containing a saturated KOH solution, 300 mL/container) to completely remove CO_2 . Secondly, the CO_2 -free air was dehumidified by passing through a container (#4, containing 200 mL of H_2SO_4). Then, CO_2 -released from the banana (in #5 container) was absorbed in two containers (#6, containing 300 mL of 0.2 M $\text{Ba}(\text{OH})_2$ with drops of phenolphthalein as a color indicator). This measurement proceeded for 60 min. The generated CO_2 was determined by titrating the remaining $\text{Ba}(\text{OH})_2$ solution (10 mL) against HCl, and respiration rate ($\text{mL CO}_2/(\text{kg sample} \times \text{h})$) was calculated by a formula:

$$\text{Respiration rate} = \frac{30 \times (3V_d - \sum V_c) \times C_M \times M_{\text{CO}_2}}{2 \times m \times d \times 10^{-3} \times t}$$

where: V_d – volume of 0.1 M HCl (mL) used for titration without sample in #5 container, V_c – volume of HCl (mL) used for titration with sample in #5 container, C_M – concentration of HCl solution (M), M_{CO_2} molecular weight of CO_2 (g/mol),

m – mass of sample (g), d – density of CO_2 (kg/kL), t – time of respiration measurement (h).

Weight loss determination

Banana weight loss was estimated during storage by a 2-digit balance (CP2P-F, Sartorius Ltd., Göttingen, Germany). The weight loss (%) was calculated following a previous study [Soradech *et al.*, 2017]:

$$\text{Weight loss} = \frac{W_0 - W_1}{W} \times 100$$

where: W_0 – weight of sample (g) at 0 day (day of storage) and W_1 – weight of banana (g) taken on each interval of storage.

Determination of soluble solid content and titratable acidity

Soluble solid content (SSC) and titratable acidity (TA) of bananas were determined during storage following methods described by Mehdi *et al.* [2011]. Banana fruit pulp (10 g) and distilled water (40 mL) were blended well. The mixture was then centrifuged at $5000 \times g$ for 5 min and then filtered through a Whatman filter paper. After that, the supernatant was measured by a refractometer (Atago Co., Ltd., Tokyo, Japan) to determine SSC as degree Brix ($^\circ\text{Bx}$). In addition, the supernatant (5.0 mL) was titrated by 0.1 M NaOH with drops of phenolphthalein as a color indicator to determine TA. The result was expressed as g of malic acid equivalent per 100 g of banana pulp fresh weight (FW).

Determination of total carbohydrate and reducing sugar contents

To determine contents of total carbohydrates and reducing sugars of stored bananas, banana pulp (2.0 g) was hydrolyzed by 2.5 M HCl (5.0 mL) for 3 h in a boiling water bath and cooled to room temperature. The mixture was then neutralized by Na_2CO_3 until the effervescence ceases. Distilled water was added to this mixture for making up to 100 mL, and the mixture was centrifuged at $12,000 \times g$ for 15 min. Total carbohydrate content was determined by the phenol-sulfuric acid method [DuBois *et al.*, 1956]. Absorbance readings were taken at 490 nm using a spectrophotometer (Halo VIS-20, Dynamica Scientific Ltd., UK). Glucose was used as a standard and total carbohydrate content was calculated using a standard curve. Results were expressed as g glucose equivalent per 100 g of banana pulp FW.

Reducing sugar content in supernatant was estimated using dinitrosalicylic acid (DNS) method [Horwitz, 1960]. Results were expressed in mg glucose/g banana pulp, based on the fresh matter.

Starch content

The starch content of stored bananas was determined by the iodine dipping method [Blankenship *et al.*, 1993]. The cross-section of the banana was dyed with an iodine solution (1% KI with 0.1% I_2 in distilled water). The sample was immersed in I_2/KI solution (5 mm of depth, 30 s). Then, the color change (dark blue) was visually observed and scored

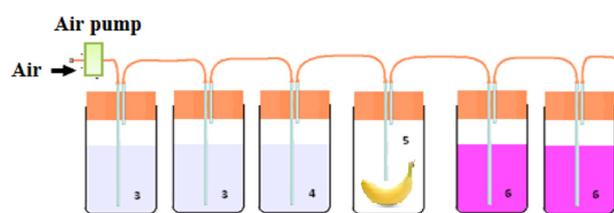


FIGURE 1. A closed respirometer for the respiration rate measurement.

Description of containers is provided in the “Respiration rate analysis subsection” of the “Materials & Method” section.

[Thakur *et al.*, 2019]. The image of color change was estimated at the rate of black-blue point using the ImageJ software (Wayne Rasband, National Institutes of Health, Bethesda, MD, USA). The results were expressed as percentage ratio (%) of black-blue area (starch) to cross-section area.

Statistical analysis

Each measurement was made for 20 banana fruits and was repeated 3 times. Data were statistically analyzed by using one-way analysis of variance (ANOVA). The significance level at $p < 0.05$ was used throughout the study. The results were ranked according to Fisher’s least significant difference (LSD) test using Statgraphics statistical analysis software version 15 (Statgraphics Technologies, Inc., The Plains, VA, US).

RESULTS AND DISCUSSION

Coating layers microstructure

The SEM micrographs of the coating layer of bananas are presented in Figure 2. The coating cross-section showed that the G0 sample had a dense and homogeneous polymer network with no pores. The results indicated that the starch granules were totally disrupted and well mixed with the plasticizers in the polymer network during the heating process. This structure promoted the barriers of gases and moisture [Chiumarelli & Hubinger, 2014]. Basiak *et al.* [2017] observed that native starch-based films had a heterogeneous structure, which resembled a fiber network. Ávila-Martín *et al.* [2020] also reported that the microstructure of achira starch-based film presented the cracks in the structure. The differences compared to electrolyzed starch coating layer in our study could be explained by the attribution of plasticizers, which formed strong bonds in the amorphous phase of starch due to its less ordered structure. Thus, the integrity of the coating was maintained by preventing the formation of pores or cracks [Basiak *et al.*, 2018].

Furthermore, as it is shown in Figure 2, adding gelatin thickened the coating layer without causing phase separation. The presence of phase separation and cracks in the structure was observed in gelatin-starch film and gelatin-glycerol film [Al-Hassan & Norziah, 2012; De Carvalho & Grosso, 2004; Moreno *et al.*, 2017]. The authors stated the heterogeneous structure of coating reduced its mechanical properties while increasing gas permeability. Interestingly, Acosta *et al.* [2015] worked on gelatin-cassava starch film and they observed the heterogeneous structures and fibrous regions.

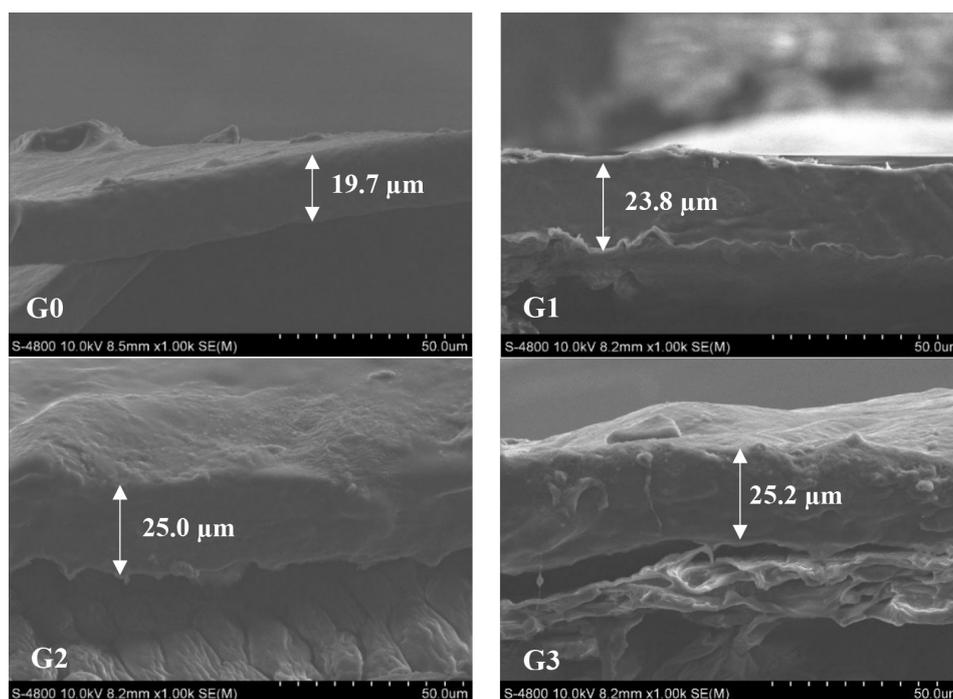


FIGURE 2. Scanning electron micrographs of coating layers of bananas coated in solutions of electrolyzed cassava starch (G0) and electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively).

Phase separation occurred even though the hydrophilic properties were similar. However, these results contrasted with the homogeneous structure of electrolyzed cassava starch-gelatin coating layer shown in Figure 2. Thus, starch oxidation by the electrolysis seems to be effective in enhancing the interaction between starch with the gelatin and plasticizers. Theoretically, coating thickness was an important parameter affecting the permeability [Cisneros-Zevallos & Krochta, 2006]. The average thickness of the G0 sample was the lowest ($19.7 \mu\text{m}$) whilst G1, G2 and G3 samples were about 23.8 , 25.0 and $25.2 \mu\text{m}$, respectively. The orientation of the starch molecules as well as the compatibility of the polymers, could explain the differences in thickness [Kumar *et al.*, 2019]. Moreover, the increase of gelatin content caused the increase in dry mass and viscosity of the solution [Fakhouri *et al.*, 2015].

Respiration rate

Respiration rate was an important indicator of fruit shelf life [Mehdi *et al.*, 2011]. In climacteric fruits, the rapid increase in the respiration rate along with a strong metabolism was an indicator of the ripening process [Wills & John, 2016].

The respiration rate of the control sample reached its peak on the 3rd day of storage (Figure 3). Interestingly, the use of coatings consisting of G0, G1, G3, and G4 samples delayed the peak of respiration until the 5th, 6th, 6th and 7th day of storage, respectively. This result was in agreement with the SEM results (Figure 3) showing the homogeneous structure of the coating layer, which limited gas dispersion [Khalil *et al.*, 2019]. G0 sample delayed the respiration rate by up to 2 days compared to control sample (Figure 3). This could be explained by the formation of cross-linking in the coating layer that reduced the presence of free hydroxyl groups, resulting in an improved gas barrier [Lawal *et al.*, 2005].

Besides, the addition of gelatin showed a benefit in reducing the ripening rate of bananas. According to Mohsen *et al.* [2017], gelatin was a good barrier to oxygen transfer. This could be explained by a crosslinking action and close packing of the components resulting from the mixing of starch-protein [Jagannath *et al.*, 2003]. It could be a small percentage of the starch and protein in the amorphous phase transformed to the crystalline phase, which was resistant to oxygen transfer in the biopolymer [Shen *et al.*, 2010]. Thus, a coating made from electrolyzed cassava starch and gelatin was strongly affected by the respiration rate of the banana. In general, climacteric fruit started the ripening by a rapid rise in respiration rate. However, when the amount of oxygen in the atmosphere falls to 2%, the fermentation occurs instead of respiration, causing unpleasant flavors and premature aging. Besides, the failure of fruit ripeness caused not only bad flavor but also internal spoilage [Emragi *et al.*, 2022]. This phenomenon was not observed in our treated samples, indicating that the coating kept appropriate low inner O_2 and high CO_2 partial pressures for fruit preservation.

Weight loss

The weight loss is related to water evaporation combined with the loss of water retention, resulting in the wrinkling of fruit [Gol & Rao, 2014]. In addition, respiratory activity also contributed to the reduction in the fruit mass due to the breakdown of sugars, such as glucose, into CO_2 and water [Santi & Jung, 1992].

It was shown in Figure 4 that the treated samples had lower weight loss than the control samples up to 5 days of storage ($p < 0.05$). Generally, the results claimed that the coating reduced gas permeability in fruit surfaces, resulting in a reduction of respiration rate, which could account for

the lower weight loss. This result is in agreement with the research of Chiumarelli & Hubinger [2014], in which the edible coatings could prevent fruits from dehydration due to their moisture barrier. There was a significant difference ($p < 0.05$) in weight loss between the control and treated samples at the beginning of storage, but the G0, G1 and G2 samples showed no significant difference ($p \geq 0.05$) with the control samples from the 6th day of storage, although the values for G3 still were significantly lower than for the control (Figure 4). The weight loss was lower by increasing the content of gelatin in the coating formula and this result was in agreement with Gol & Rao [2014] and Aguilar-Méndez *et al.* [2008]. Thereby, this coating could reduce moisture loss and prolong the shelf life of fruit.

Soluble solid content

The SSC of control, electrolyzed starch and electrolyzed starch-gelatin coated banana fruits during storage is shown in Figure 5. The SSC increased along with fruit ripening. It was probably due to respiration rate, degradation of polysaccharides, or water evaporation [Moreira *et al.*, 2021]. After 8 days of storage, the SSC of the control sample was higher compared with the coated samples (Figure 5). Generally, SSC of coated samples was lower than that of the control during ripening because the hydrolysis of starches and carbohydrates into soluble sugars was delayed [Dwivany *et al.*, 2020].

A layer of coating on the surface of the fruit changed the intrinsic atmosphere and concentration of O_2 and CO_2 . In other words, the coating delayed the ripening reflected by lower SSC of banana fruits. For the coated samples, the SSC of the G3 sample was the lowest from the 4th day of storage (Figure 5). Obviously, the content of gelatin had a significant impact on the SSC of the samples. SSC decreased when the amount of gelatin in the coating formula increased. This result was in agreement with Moreira *et al.* [2021], who reported gelatin-starch coated guavas had lower SSC during storage than the control sample.

Titrateable acidity

The organic acids were oxidized by the respiration process in climacteric fruits. As a result, the TA was decreased during storage time [Moreira *et al.*, 2021]. The results showed that TA of the samples increased gradually until it reached the peak and then sharply decreased with fruit maturation (Figure 6).

Many organic acids such as malic (a principle acid), citric, oxalic, and tartaric acids are present in banana fruit. Oxalic acid caused astringent taste whilst malic and citric contributed to the tartness of banana fruits [Seymour, 1993]. The content of these acids decreased during ripening due to their use as substrates for respiratory enzyme reactions [Seymour, 1993]. This phenomenon may have occurred in our experiment since TA and respiration correlated significantly (Figure 3 and Figure 6). The results agreed with the finding of Márquez Cardozo *et al.* [2015] showing that TA was related to the respiratory rate. Interestingly, the coating showed a reduction in the respiration rate, therefore delaying the decline in acidity. Furthermore, the higher the gelatin content in the coating solution was, the longer it took to reach the TA peak and the lower the TA reduction of banana fruits (Figure 6).

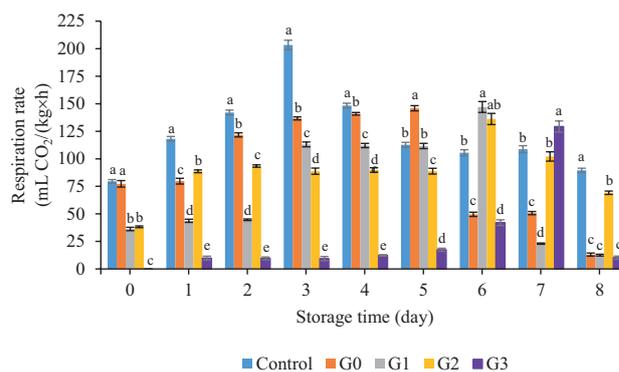


FIGURE 3. Respiration rate of bananas coated in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

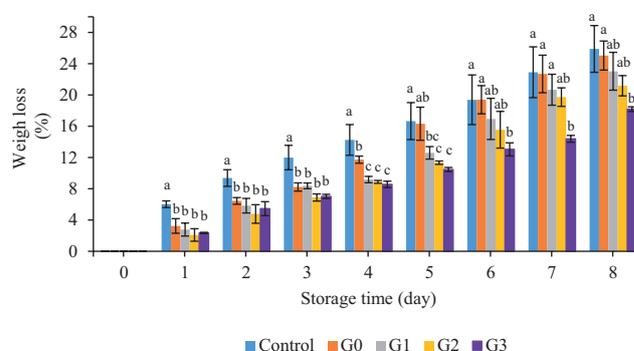


FIGURE 4. Weight loss of bananas coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

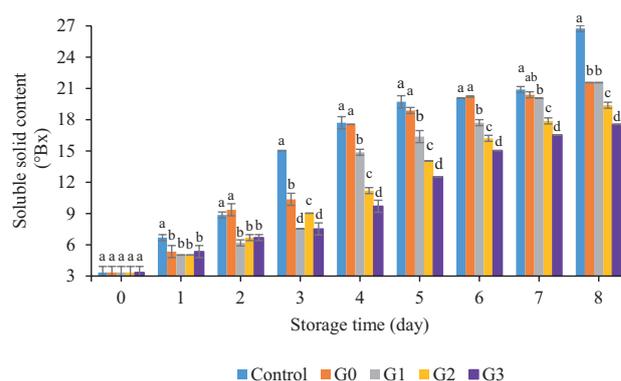


FIGURE 5. Soluble solid content of bananas coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

If gelatin is added to the coating, it bonds together to form a gelatin network, reduces the holes for air to pass through, forms a denser polymeric matrix and increases the resistance to gas permeability [Fakhouri *et al.*, 2012].

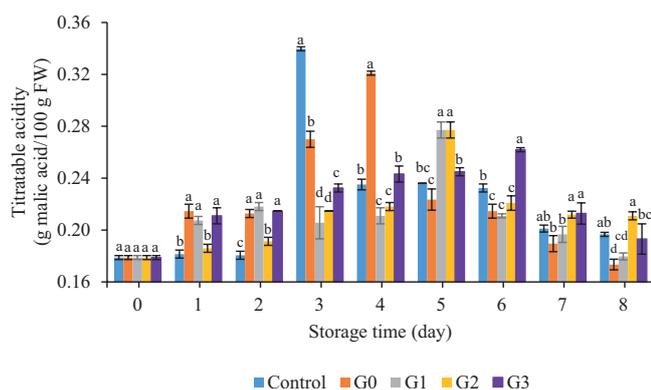


FIGURE 6. Titrateable acidity of bananas coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. FW – fresh weight. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

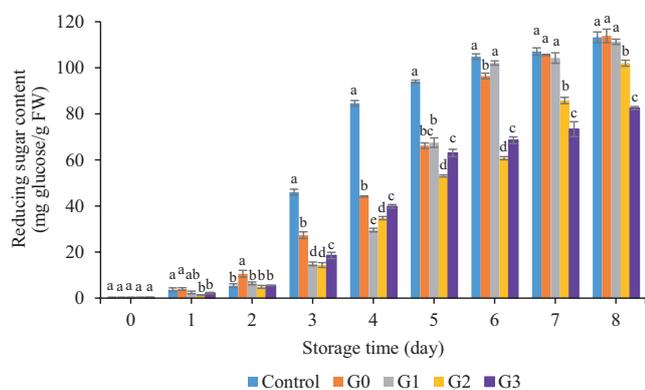


FIGURE 7. Reducing sugar content of bananas coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. FW – fresh weight. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

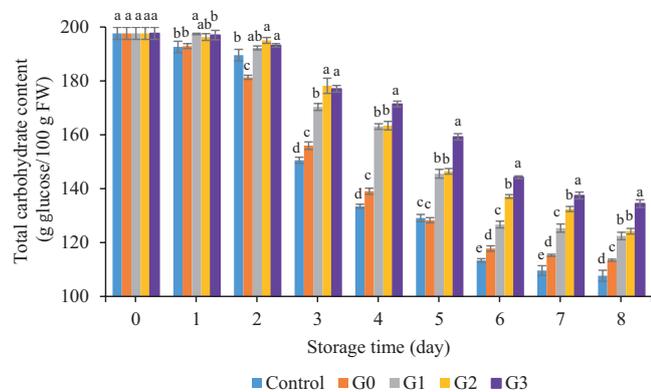


FIGURE 8. Total carbohydrate content of bananas coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) stored for 8 days. FW – fresh weight. Different letters above bars on the same day of storage indicate a statistically significant difference, $p < 0.05$.

Reducing sugar and total carbohydrate contents

After harvesting, reducing sugar content of unripe banana fruit was very low (0.3 mg glucose/g FW). However, the content of reducing sugar raised to 80–111 mg glucose/g FW when the fruit ripened (Figure 7). Reducing sugars in ripened bananas were mainly glucose and fructose [Maduwanthi & Marapana, 2017]. The activity of amylase in banana led to the hydrolysis of starch into soluble sugars. In this study, reducing sugar contents were ordered as: control=G0=G1>G2>G3 after 8 day of storage. This result was due to the effect of the coating, which contributed to the reduction of the respiration rate and ethylene production, resulting in the starch hydrolysis taking place slowly [Thakur *et al.*, 2019]. Additionally, increasing the ratio of gelatin in coating composition was beneficial in reducing the content of reducing sugars and carbohydrates. This finding was supported by Gol & Rao [2014] study, which found that gelatin-based coatings were more effective at delaying ripening and maintaining fruit quality.

Total carbohydrate content in all samples tended to decrease gradually during 8 days of storage (Figure 8). The total carbohydrate contents at 8th-day storage were sorted as: control<G0<G1=G2<G3. Interestingly, in this study, the carbohydrate contents were negatively correlated to the maximal value of respiration rates. During ripening, starch and pectin were degraded. Especially, starch was hydrolyzed to simple sugars which are the main substrates for respiration [Gol & Rao, 2014]. A rapid increase in the respiration rate in banana fruits results from the loss of starch and sugar. Actually, the respiration process converted glucose into CO₂ gas [Elhadi *et al.*, 2019]. Thus, these phenomena were good explanations for the loss of total carbohydrates in our study. This result was consistent with the respiration rate results shown in Figure 3, where the coatings reduced the fruit respiratory rate, and the effect was enhanced with the addition of gelatin.

Starch content

The starch content of all banana samples was very high during the first days of storage (Figure 9). The persistence of a blue/black coloration after dyed with an iodine solution in the coated fruit compared to the control indicates delayed ripening. During the ripening, it sharply decreased until no starch was detected in the fruit. The rate of starch breakdown was much greater in control fruits ($p < 0.05$) where 100% starch degradation was recorded at the end of day 6 (no blue/black color). However, at the same storage time, the values were, 16.1%, 38.9%, 44.3%, 88.6% for G0, G1, G2 and G3 sample, respectively. Interestingly, the coated samples (G1, G2, G3) still had the presence of starch in the banana pulp after 8 days of storage.

Most of the carbohydrates in unripe bananas were in the form of starch, which was hydrolyzed into sucrose then glucose and fructose during the short period of ripening [Gol & Rao, 2014]. The conversion of starch into sucrose during ripening was a complex process but the main pathway for starch degradation was described by Terra *et al.* [1983]. Starch content was extremely high in unripe fruit. It was rapidly reduced as a result of respiration, down to less than 1%. On the other hand, sucrose gradually increased until reaching its peak.

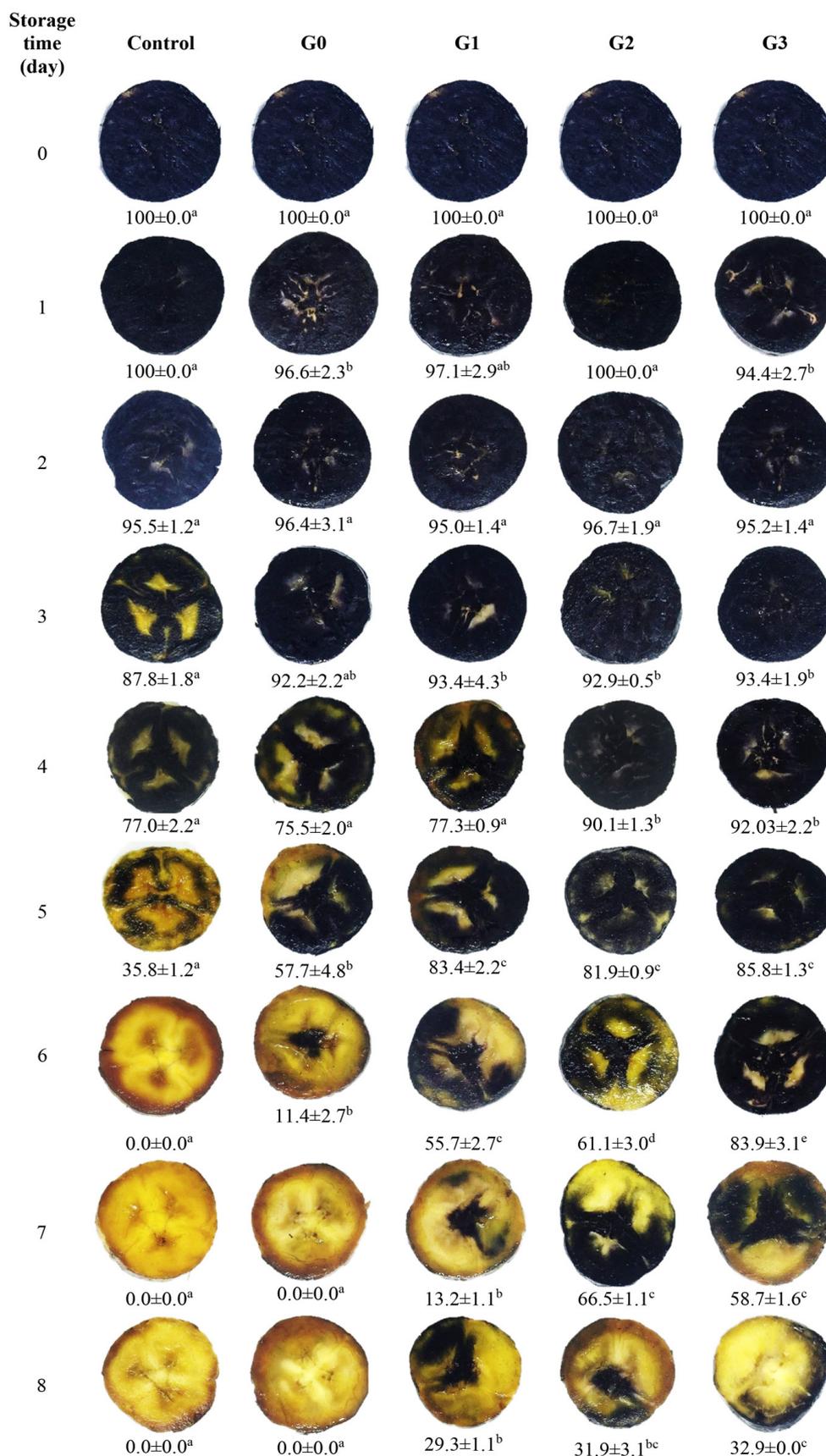


FIGURE 9. Images of the cross-section of the bananas after dyeing with an iodine solution and starch content in bananas (% of black-blue area (starch) to cross-section area). Bananas were coated by dipping in solutions of electrolyzed cassava starch (G0), electrolyzed cassava starch with 1, 2 and 3% (w/v) gelatin (G1, G2 and G3, respectively) and uncoated fruits (control) and stored for 8 days.

Data were shown as mean ± standard deviation; different letters in the same row indicate a statistically significant difference, $p < 0.05$.

Moreover, the images showed that starch began to degrade in the central core of banana pulp and toward the outside during the ripening (Figure 9). It could be observed that the initial active enzyme area was not random due to the higher sensitivity of interior starch granule to degradation by enzyme [Terra *et al.*, 1983]. The breakdown of starch into sugar was more clearly observed at the cross-section of banana fruit. This was due to a sharp increase of ethylene on the pulp-peel interface [Yun *et al.*, 2019]. This finding demonstrated that the semi-anaerobic environment formed by surface coating decreased the activity of starch degrading enzyme and the effectiveness was enhanced with increasing concentration of gelatin.

CONCLUSION

In this study, the coating made from electrolyzed cassava starch and gelatin was effective in delaying banana ripening. The coating formulation showed good resistance to gas migration. The addition of gelatin in oxidized starch coating enhanced its ability to diminish respiratory intensity. Overall, the coating treatment with gelatin 3% (w/v) proved the most effective when it could prolong shelf life more than 2 times compared to the control sample. The electrolyzed cassava starch-gelatin coating could be a potential material to increase the shelf life of banana as well as other climacteric fruits.

RESEARCH FUNDING

The author(s) received no financial support for the research, authorship, and/or publication of this article.

ACKNOWLEDGMENTS

The authors acknowledge the Doctoral School of Food Science of the Hungarian University of Agriculture and Life Sciences for the support in this study. Special gratitude is given to HCMC University of Technology and Education for stimulating suggestions and encouragement that helped us to finish this study.

CONFLICTS OF INTERESTS

The authors declare that they have no conflicts of interest.

DATA AVAILABILITY STATEMENT

The data used to support the findings of this study are available from the corresponding author upon request.

ORCID IDs

T.T. Pham <https://orcid.org/0000-0002-7911-6430>

K.S. Trinh <https://orcid.org/0000-0002-6365-2693>

REFERENCES

1. Acosta, S., Jiménez, A., Cháfer, M., González-Martínez, C., Chiralt, A. (2015). Physical properties and stability of starch-gelatin

based films as affected by the addition of esters of fatty acids. *Food Hydrocolloids*, 49, 135–143.

<https://doi.org/10.1016/j.foodhyd.2015.03.015>

2. Agarwal, S. (2021). Major factors affecting the characteristics of starch based biopolymer films. *European Polymer Journal*, 160, art. no. 110788.

<https://doi.org/10.1016/j.eurpolymj.2021.110788>

3. Aguilar-Méndez, M.A., Martín-Martínez, E.S., Tomás, S.A., Cruz-Orea, A., Jaime-Fonseca, M.R. (2008). Gelatine–starch films: Physicochemical properties and their application in extending the post-harvest shelf life of avocado (*Persea americana*). *Journal of the Science of Food and Agriculture*, 88(2), 185–193.

<https://doi.org/10.1002/jsfa.3068>

4. Al-Hassan, A.A., Norziah, M.H. (2012). Starch–gelatin edible films: Water vapor permeability and mechanical properties as affected by plasticizers. *Food Hydrocolloids*, 26(1), 108–117.

<https://doi.org/10.1016/j.foodhyd.2011.04.015>

5. Ávila-Martín, L., Beltrán-Osuna, Á.A., Perilla, J.E. (2020). Effect of the addition of citric acid and whey protein isolate in *Canna indica* L. starch films obtained by solvent casting. *Journal of Polymers and the Environment*, 28, 871–883.

<https://doi.org/10.1007/s10924-019-01648-z>

6. Bailey C.H. (1940). Respiration of cereal grains and flaxseed. *Plant Physiology*, 15(2), 257–274.

<https://doi.org/10.1104/pp.15.2.257>

7. Basiak, E., Lenart, A., Debeaufort, F. (2017). Effect of starch type on the physico-chemical properties of edible films. *International Journal of Biological Macromolecules*, 98, 348–356.

<https://doi.org/10.1016/j.ijbiomac.2017.01.122>

8. Basiak, E., Lenart, A., Debeaufort, F. (2018). How glycerol and water contents affect the structural and functional properties of starch-based edible films. *Polymers*, 10(4), art. no. 412.

<https://doi.org/10.3390/polym10040412>

9. Blankenship, S., Ellsworth, D.D., Powell, R.L. (1993). A ripening index for banana fruit based on starch content. *American Society for Horticultural Science*, 3(3), 338–339.

<https://doi.org/10.21273/HORTTECH.3.3.338>

10. Chiumarelli, M., Hubinger, M.D. (2014). Evaluation of edible films and coatings formulated with cassava starch, glycerol, carnauba wax and stearic acid. *Food Hydrocolloids*, 38, 20–27.

<https://doi.org/10.1016/j.foodhyd.2013.11.013>

11. Cisneros-Zevallos L., Krochta J.M. (2006). Dependence of coating thickness on viscosity of coating solution applied to fruits and vegetables by dipping method. *Food Science*, 68(2), 503–510.

<https://doi.org/10.1111/j.1365-2621.2003.tb05702.x>

12. Cortés-Rodríguez, M., Villegas-Yépez, C., González, J.H.G., Rodríguez, P.E., Ortega-Toro, R. (2020). Development and evaluation of edible films based on cassava starch, whey protein, and bees wax. *Heliyon*, 6(9), art. no. e04884.

<https://doi.org/10.1016/j.heliyon.2020.e04884>

13. De Carvalho, R.A., Grosso, C.R.F. (2004). Characterization of gelatin based films modified with transglutaminase, glyoxal and formaldehyde. *Food Hydrocolloids*, 18(5), 717–726.

<https://doi.org/10.1016/j.foodhyd.2003.10.005>

14. DuBois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A., Smith, F. (1956). Colorimetric method for determination of sugars and related substances. *Analytical Chemistry*, 28(3), 350–356.

<https://doi.org/10.1021/ac60111a017>

15. Dwivany, F.M., Aprilyandi, A.N., Suendo, V., Sukriandi, N. (2020). Carrageenan edible coating application prolongs caven-dish banana shelf life. *International Journal of Food Science*, 2020, art. no. 8861610.
<https://doi.org/10.1155/2020/8861610>
16. Elhadi M.Y., Carillo-Lopez, A., Bello-Perez, A.A. (2019). Carbo-hydrates. Chapter 9. In E. Yahia, A. Carrillo-Lopez (Eds.). *Post-harvest Physiology and Biochemistry of Fruits and Vegetables*. 2nd edition, Woodhead Publishing: Elsevier Inc.
17. Emragi, E., Kalita, D., Jayanty, S.S. (2022). Effect of edible coat-ing on physical and chemical properties of potato tubers under different storage conditions. *LWT – Food Science and Technology*, 153, art. no. 112580.
<https://doi.org/10.1016/j.lwt.2021.112580>
18. Fakhouri, F.M., Martelli, S.M., Caon, T., Velasco, J.I., Mei, L.H.I. (2015). Edible films and coatings based on starch/gelatin: Film properties and effect of coatings on quality of refrigerated Red Crimson grapes. *Postharvest Biology and Technology*, 109, 57–64.
<https://doi.org/10.1016/j.postharvbio.2015.05.015>
19. Fakhouri, F.M., Martelli, S.M., Bertan, L.C., Yamashita, F., Mei, L.H.I., Queiroz, F.P.C. (2012). Edible films made from blends of manioc starch and gelatin – Influence of different types of plasticizer and different levels of macromolecules on their properties. *LWT – Food Science and Technology*, 49(1), 149–154.
<https://doi.org/10.1016/j.lwt.2012.04.017>
20. FAO. (2021). *Banana Statistical Compendium 2020*. Rome.
21. Gol, N.B., Rao, T.R. (2014). Influence of zein and gelatin coat-ings on the postharvest quality and shelf life extension of mango (*Mangifera indica* L.). *Fruits*, 69(2), 101–115.
<https://doi.org/10.1051/fruits/2014002>
22. Horwitz, W. (1960). Use of dinitrosalicylic acid reagent for deter-mination of reducing sugar. *Official Methods of Analysis of the As-sociation of Official Analytical Chemists*. 9th ed. Washington DC: Association of Official Analytical Chemists.
23. Hossain, M.S., Iqbal, A. (2016). Effect of shrimp chitosan coat-ing on postharvest quality of banana (*Musa sapientum* L.) fruits. *International Food Research Journal*, 23(1), 277–283.
24. Jagannath, J.H., Nanjappa, C., Das Gupta, D.K., Bawa, A.S. (2003). Mechanical and barrier properties of edible starch–pro-tein-based films. *Journal of Applied Polymer Science*, 88(1), 64–71.
<https://doi.org/10.1002/app.11602>
25. Kader, A.A. (2005). *Banana respiration, in Banana, Recommendations for Maintaining Postharvest Quality*. Postharvest Technology Research Information Centre, UCDAVIS: Davis, CA, USA.
26. Karaca, H., Pérez-Gago, M.B., Taberner, V., Palou, L. (2014). Evaluating food additives as antifungal agents against *Mon-illinia fructicola* in vitro and in hydroxypropyl methylcellulose-lipid composite edible coatings for plums. *International Journal of Food Microbiology*, 179, 72–79.
<https://doi.org/10.1016/j.ijfoodmicro.2014.03.027>
27. Khalil, H.A., Saurabh, C.K., Syakir, M.I., Fazita, M.N., Bhat, A., Banerjee, A., Fizree, H.M., Rizal, S., Tahir, P.M. (2019). Barrier properties of biocomposites/hybrid films. In *Mechanical and Physical Testing of Biocomposites, Fibre-Reinforced Compos-ites and Hybrid Composites*, Woodhead Publishing, pp. 241–258.
<https://doi.org/10.1016/B978-0-08-102292-4.00013-8>
28. Kim, S.R.B., Choi, Y.G., Kim, J.Y., Lim, S.T. (2015). Im-provement of water solubility and humidity stability of tapioca starch film by incorporating various gums. *LWT – Food Science and Technology*, 64(1), 475–482.
<https://doi.org/10.1016/j.lwt.2015.05.009>
29. Kokoszka, S., Lenart, A. (2007). Edible coatings-formation, characteristics and use-a review. *Polish Journal of Food and Nutri-tion Sciences*, 57(4), 399–404.
30. Kudachikar, V.B., Kulkarni, S.G., Prakash, M.K. (2011). Ef-fect of modified atmosphere packaging on quality and shelf life of ‘Robusta’ banana (*Musa* sp.) stored at low temperature. *Jour-nal of Food Science and Technology*, 48(3), 319–324.
<https://doi.org/10.1007/s13197-011-0238-y>
31. Kumar, R., Ghoshal, G., Goyal, M. (2019). Synthesis and func-tional properties of gelatin/CA–starch composite film: excellent food packaging material. *Journal of Food Science and Technology*, 56(4), 1954–1965.
<https://doi.org/10.1007/s13197-019-03662-4>
32. Lawal, O.S., Adebowale, K.O., Ogunsanwo, B.M., Barba, L.L., Ilo, N.S. (2005). Oxidized and acid thinned starch derivatives of hybrid maize: functional characteristics wide-angle X-ray dif-fractometry and thermal properties. *International Journal of Bio-logical Macromolecules*, 35(1–2), 71–79.
<https://doi.org/10.1016/j.ijbiomac.2004.12.004>
33. Le Nguyen, L.P., Visy, A., Baranyai, L. (2020). Application of hue spectra fingerprinting during cold storage and shelf-life of pack-aged sweet cherry. *Food Measure*, 14, 2689–2702.
<https://doi.org/10.1007/s11694-020-00515-z>
34. Maduwanthi, S.D.T., Marapana, R.A.U.J. (2017). Biochemical changes during ripening of banana: A review. *International Jour-nal of Food Science and Nutrition*, 2(5), 166–169.
35. Márquez Cardozo, C.J., Palacín Beltrán, J.R., Fuentes Berrio, L. (2015). Effect of cassava-starch coatings with ascorbic acidic and N-acetylcysteine on the quality of harton plantain (*Musa paradisiaca*). *Revista Facultad Nacional de Agronomía Medellín*, 68(2), 7689–7701.
<https://doi.org/10.15446/rfnam.v68n2.50985>
36. Mehdi, M., Asgar A., Peter, G.A., Noosheen, Z., Yasmeen, S. (2011). Effect of a novel edible composite coating based on gum arabic and chitosan on biochemical and physiological respons-es of banana fruits during cold storage. *Journal of Agricultural and Food Chemistry*, 59(10), 5474–5482.
<https://doi.org/10.1021/jf200623m>
37. Mohsen, R.E.F., Hamidreza, A., Sedigheh, A. (2017). Effect of gelatin-based edible coatings incorporated with *Aloe vera* and Black and Green tea extracts on the shelf life of fresh-cut oranges. *Journal of Food Quality*, 2017, art. no. 9764650.
<https://doi.org/10.1155/2017/9764650>
38. Moreira, E.D.S., Silva, N.M.C.D., Brandão, M.R.S., Santos, H.C., Ferreira, T.A.P.D.C. (2021). Effect of modified starch and gelatin by-product based edible coating on the postharvest quality and shelf life of guava fruits. *Food Science and Technology*, 42, art. no. e26221.
<https://doi.org/10.1590/fst.26221>
39. Moreno, O., Cárdenas, J., Atarés, L., Chiralt, A. (2017). Infl-uence of starch oxidation on the functionality of starch-gelatin based active films. *Carbohydrate Polymers*, 178, 147–158.
<https://doi.org/10.1016/j.carbpol.2017.08.128>
40. Neelam, P., Mehar, H.A., Puneet, D., Manoj, K.S., Pravendra, N. (2003). Expression and activities of ethylene biosynthesis

- enzymes during ripening of banana fruits and effect of 1-MCP treatment. *Plant Growth Regulation*, 40(1), 11–19.
<https://doi.org/10.1023/A:1023040812205>
41. Podshivalov, A., Toropova, A., Fokina, M., Uspenskaya, M. (2020). Surface morphology formation of edible holographic marker on potato starch with gelatin or agar thin coatings. *Polymers*, 12(5), art. no. 1123.
<https://doi.org/10.3390/polym12051123>
42. Podshivalov, A., Zakharova, M., Glazacheva, E., Uspenskaya, M. (2017). Gelatin/potato starch edible biocomposite films: Correlation between morphology and physical properties. *Carbohydrate Polymers*, 157, 1162–1172.
<https://doi.org/10.1016/j.carbpol.2016.10.079>
43. Pranoto, Y., Paramita, B.L., Cahyanto, M.N., Benjakul, S. (2021). Properties of ozone-oxidized tapioca starch and its use in coating of fried peanuts. *Molecules*, 26(20), art. no. 6281.
<https://doi.org/10.3390/molecules26206281>
44. Radziejewska-Kubzdela, E., Bieganska-Marecik, R., Czapski, J. (2007). Effect of composition of atmosphere inside packaging on quality of minimally processed vegetable salad based on white cabbage. *Polish Journal of Food and Nutrition Sciences*, 57(4C), 461–465.
45. Salehi, F. (2020). Edible coating of fruits and vegetables using natural gums: A review. *International Journal of Fruit Science*, 20(sup2), S570–S589.
<https://doi.org/10.1080/15538362.2020.1746730>
46. Santi, R.B., Jung, C.P. (1992). Shelf-life of mature green tomatoes stored in controlled atmosphere and high humidity. *Journal of Food Science*, 57(4), 948–953.
<https://doi.org/10.1111/j.1365-2621.1992.tb14331.x>
47. Seymour G.B. (1993). *Banana. Biochemistry of Fruit Ripening*. Springer Netherlands, pp. 83–106.
<https://doi.org/10.1007/978-94-011-1584-1>
48. Shen, X.L., Wu, J.M., Chen, Y., Zhao, G. (2010). Antimicrobial and physical properties of sweet potato starch films incorporated with potassium sorbate or chitosan. *Food Hydrocolloids*, 24(4), 285–290.
<https://doi.org/10.1016/j.foodhyd.2009.10.003>
49. Soradech, S., Nunthanid, J., Limmatvapirat, S., Luangtana-anan, M. (2017). Utilization of shellac and gelatin composite film for coating to extend the shelf life of banana. *Food Control*, 73, 1310–1317.
<https://doi.org/10.1016/j.foodcont.2016.10.059>
50. Terra, N.N., Garcia, E., Lajolo, F.M. (1983). Starch-sugar transformation during banana ripening: the behavior of UDP glucose pyrophosphorylase, sucrose synthetase and invertase. *Journal of Food Science*, 48(4), 1097–1100.
<https://doi.org/10.1111/j.1365-2621.1983.tb09169.x>
51. Thakur, R., Pristijono, P., Bowyer, M., Singh, S.P., Scarlett, C.J., Stathopoulos, C.E., Vuong, Q.V. (2019). A starch edible surface coating delays banana fruit ripening. *LWT – Food Science and Technology*, 100, 341–347.
<https://doi.org/10.1016/j.lwt.2018.10.055>
52. Trinh, K.S., Dang, T.B. (2019). Structural, physicochemical, and functional properties of electrolyzed cassava starch. *International Journal of Food Science*, 2019, art. no. 9290627.
<https://doi.org/10.1155/2019/9290627>
53. Wills, R.B.H., John, B.G. (2016). *Physiology and biochemistry. Postharvest: An Introduction to the Physiology and Handling of Fruit and Vegetables*. 6th edition, NewSouth Publishing, pp. 34–63.
<https://doi.org/10.1079/9781786391483.0000>
54. Yun, Z., Li, T., Gao, H., Zhu, H., Gupta, V.K., Jiang, Y., Duan, X. (2019). Integrated transcriptomic, proteomic, and metabolomics analysis reveals peel ripening of harvested banana under natural condition. *Biomolecules*, 9(5), art. no. 167.
<https://doi.org/10.3390/biom9050167>
55. Zsom, T., Strohmayer, E., Phuong Le Nguyen, L., Hitka, G., Zsom-Muha, V. (2018). Chilling injury investigation by non-destructive measuring methods during banana cold storage. *Progress in Agricultural Engineering Sciences*, 14(S1), 147–158.
<https://doi.org/10.1556/446.14.2018.s1.14>