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Pectin-Based Edible Coating Combined with Chemical Dips Containing Antimicrobials and Antibrowning Agents to Maintain Quality of Fresh-Cut Pears

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Abstract: The aim of this study was to assess the effects of pectin coating alone (PE) or combined with chemical dips containing potassium sorbate (PS) or sodium benzoate (SB) as antimicrobials, and *N*-acetyl cysteine (*N*-AC) or ascorbic acid (AA) + citric acid (CA) as antibrowning agents, on weight loss, color values, browning index, firmness, titratable acidity, soluble solids content, total phenolic content, antioxidant activity and sensory attributes of fresh-cut pears during 15-day storage at 8 °C. Pectin coating delayed weight loss and improved firmness of fresh-cut pears as compared to control samples. Addition of either 1% *N*-AC or 1% CA + 1% AA in the formulation of the chemical dip protected the phenolic compounds and enhanced the antioxidant activity of fresh-cut pears during storage. PE + 0.2% SB + 1% *N*-AC and PE + 0.2% PS + 1% *N*-AC were the most efficient treatments in preserving color and reducing the browning index of fresh-cut pears during 15-day storage at 8 °C and received the highest scores for all sensory attributes throughout 12 days of storage. The results demonstrate the feasibility of PE + 0.2% SB + 1% *N*-AC and PE + 0.2% PS + 1% *N*-AC for extending the shelf life of fresh-cut pears.

Keywords: potassium sorbate; sodium benzoate; *N*-acetyl cysteine; ascorbic acid; citric acid; calcium chloride; storage

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1. Introduction

Consumers' recent interest in fresh, varied and more convenient foods, coupled with changes in their lifestyles, has led to an increase in the production and consumption of minimally processed foods [1]. Furthermore, as consumers are more aware of the health benefits related to the intake of fruits and vegetables, in recent years the fresh-cut fruit and vegetable market has become more significant [2]. For fruits, "minimal processing" refers to physical changes in the initial state, for example, by trimming, peeling, washing and/or cutting, so that the resulting product may be directly eaten, in a fresh state, without previous preparation [3]. The disruption of fruit tissue integrity in mechanical processing operations and larger cut surfaces increase the susceptibility of fresh-cut fruits to enzymatic browning, water loss, texture softening, intensified respiration, production of undesirable odors and flavors and microbial spoilage [4,5]. As a result, fresh-cut fruits have a much shorter shelf-life as compared to intact fruits, determined by deterioration in quality parameters such as color, firmness, juiciness and flavor [1,6]. Temperature, relative humidity, atmospheric composition and ethylene concentration are the main external factors that influence the shelf life of fresh-cut fruits [5]. In addition to changes in packaging technology, such as modified atmosphere packaging and the use of innovative packaging materials, dipping treatments and edible coatings stands as alternatives for preserving the quality and extending the shelf life of fresh-cut fruits by lowering respiration intensity, water

loss and enzymatic browning [7]. Edible coatings, applied on the fresh-cut fruit surface by dipping or spraying, act as a primary packaging and function as a semipermeable barrier that can prevent moisture loss, modify the internal gas composition, reduce respiration and oxidative reactions and delay physiological fruit ripening [8–10]. The functional properties of edible coatings can be improved by the addition of active ingredients into the polymer matrix, such as antimicrobials, texture enhancers, antibrowning agents, antioxidants and nutraceutical substances [5,6].

Edible coatings can be prepared from long-chain polysaccharides, proteins, lipids or their combinations. Due to their hydrophilic nature, proteins and polysaccharides have poor moisture barrier properties, but films and coatings made from them are good barriers against oxygen [11]. Carrageenan, carboxymethyl cellulose, pectin, alginate and chitosan are some of the polysaccharides found to be promising in obtaining edible coatings with good protective properties for fresh-cut fruits [3].

Pectin is a high-molecular-weight water-soluble polysaccharide, mainly composed of (1 → 4) linked α-d-galacturonic acid esterified units, which confer structure to the primary cell walls of plants and fruits. In the presence of calcium or other multivalent metal cations, the pectin polymer chains form a flexible network, turning into a strong gel [12,13]. Pectin is industrially extracted from various fruit by-products, such as apple pomace and citrus peels, and is added to food products with a view to increase their viscosity and gel strength [9,14]. As clearly established in previous studies, the calcium treatment needed to induce pectin gelation (usually 1% $CaCl_2$) contributes to maintaining fruit firmness during storage [15,16]. However, the residual calcium chloride on the fruit's cut surface may impart bitterness that is sometimes detected by consumers [17].

In order to increase the microbiological stability of fresh-cut fruits, the incorporation of antimicrobials such as nisin, lysozyme, organic acids and essential oils into edible coatings was proposed and tested [6]. This proved to be advantageous over the direct application of preservatives onto foods because edible coatings are able to maintain a continuous action of the antimicrobial compounds on the cut-fruit surface and to control their diffusion inside fruit [16]. Although less studied as edible coating ingredients, potassium sorbate and sodium benzoate are food additives generally recognized as safe (GRAS) and commonly added to foods as antimicrobials.

During storage, fresh-cut fruits are prone to browning, a phenomenon assigned to the action of polyphenol oxidase and other oxidative enzymes on the natural phenolic compounds released at the cut surface from the injured cells and one that adversely affects the quality and attractiveness of the product. Dipping in solutions of antibrowning agents, such as ascorbic acid and its derivatives, cinnamic acids, cysteine, glutathione, sodium chlorite, ethanol, sulfites and plant extracts, or their incorporation into edible coatings have been tested in previous work in order to inhibit or retard the browning of fresh-cut fruits [18–20]. The use of methylcellulose edible coating in combination with ascorbic acid, calcium chloride and ascorbic acid was found to prolong the shelf life of fresh-cut pears by slowing browning and strengthening texture [21]. A chemical dip containing 2% ascorbic acid + 1% calcium lactate + 0.5% cysteine significantly prevented surface browning of fresh-cut pears [22], as did alginate and gellan coatings incorporating glutathione and *N*-acetylcysteine [12]. Browning of fresh-cut apples was successfully reduced by using carrageenan and whey protein coatings incorporating ascorbic acid, citric acid or cysteine with alginate and gellan coatings containing *N*-acetylcysteine [23,24].

Pears are very popular fruits due to their appetizing and juicy flavor and their nutritional richness [25]. Pears are a good source of bioactive compounds, but their high moisture content and nutrient composition make them more susceptible to microbial contamination. Being a climacteric fruit, pears show a rise in respiration and ethylene production at the onset of ripening, which makes them more sensitive to various physiological storage-induced disorders [26].

The aim of this study was to preserve the quality of fresh-cut pears during storage at 8 °C by using pectin-based edible coatings combined with chemical dips containing

potassium sorbate (PS) or sodium benzoate (SB) as antimicrobials and either *N*-acetyl cysteine (*N*-AC) or the combination of ascorbic acid (AA) + citric acid (CA) as antibrowning agents. Weight loss, color values, browning index, firmness, titratable acidity, soluble solids content, total phenolic content, antioxidant activity and sensory attributes of the fresh-cut pears were monitored during 15 days of storage at 8 °C.

2. Materials and Methods

2.1. Chemicals

Folin–Ciocalteu reagent, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), gallic acid, sodium carbonate and 2,2-diphenyl-1-picrylhydrazyl (DPPH) were purchased from Sigma-Aldrich (Taufkirchen, Germany). Sodium benzoate and potassium sorbate were purchased from S.C. Coseli S.A. (Iași, Romania). Sosa Fruit Pectin NH (PE) (Sosa Ingredients S.L Moià, Barcelona, Spain) was used in the coating formulations, while glycerol from Merck (Darmstadt, Germany) was added to the coatings as a plasticizer. Calcium chloride (Sigma-Aldrich Chemic, Steinhein, Germany) was used to induce cross-linkage between the polymer chains. Ascorbic acid and citric acid from Merck (Darmstadt, Germany) and *N*-acetyl-cysteine from Myprotein (Manchester, UK) were used as antibrowning agents.

2.2. Plant Material

Pears (cv. Margarit, Turkey) at the commercial maturity stage were obtained from a local supermarket in November 2021. The fruits were transferred to the laboratory and were stored for 24 h at 4 °C and 85% relative humidity before processing. Fruits of similar size without diseases, physiological defects or physical damage were selected as experimental materials. They were washed with tap water, drained and placed on filter paper.

2.3. Preparation of Edible Coatings and Crosslinking Solutions

Pectin coating preparation was adapted from Moreira et al. [27]. Pectin powder was dissolved in distilled water at 2% (*w*/*v*) at a temperature of 70 °C under stirring until it became clear. After cooling to room temperature, 0.1% glycerol was added as a plasticizer. An aqueous crosslinking solution based on 1% calcium chloride was prepared. The preservatives and antibrowning agents were dissolved in the aqueous crosslinking solution. The treatment formulations and their respective codes are presented in Table 1.

Table 1. Treatment formulations and respective codes.

Treatment	Components
T0	Distilled water (control)
T1	2% PE + 1% $CaCl_2$
T2	2% PE + 1% $CaCl_2$ + 0.2% PS
T3	2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC
T4	2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA
T5	2% PE + 1% $CaCl_2$ + 0.2% SB
T6	2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC
T7	2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA

2.4. Fruit Preparation and Treatment

Pears were manually sliced transversely (20 mm thick) and cut into cuboids (2–3 cm) on a plastic board with a sharp knife, and all core tissue was removed. These cuboids were peeled before treatment and allocated to eight different treatments in a randomized manner. The freshly cut pear cuboids were first dipped for 2 min into the pectin-based film-forming solution. The excess coating solution was allowed to drip off for 1 min before a subsequent 2 min immersion into the crosslinking dip. Samples were then drained, air dried and then placed into closed transparent disposable plastic containers (750 mL capacity) and stored at 8 °C and 85% relative humidity for 15 days. Ten cuboids from different pears, weighing in

total approximately 160 g, were placed into each container, avoiding overlapping. Seven containers were prepared per treatment. The experiment was repeated three times.

Weight loss, firmness, color, total soluble solids, titratable acidity, total phenolic content and DPPH antioxidant activity were evaluated at 0, 3, 6, 9, 12 and 15 days during storage. Each determination was run in triplicate.

2.5. Weight Loss

Weight of each plastic container was measured at the start of the experiment (time 0) and at 3, 6, 9, 12 and 15 days during storage using a digital balance (Sartorius CP124S, UK, accuracy = 0.01 g). Weight loss was reported as a percentage of the initial sample weight and calculated as follows [8]:

$$\text{Weight loss\%} = (\text{Initial weight} - \text{weight in the specific storage time})/\text{initial weight} \times 100$$

2.6. Color Measurements

The CIE color values [L*(lightness), a*(redness), b*(yellowness)] of control and treated fresh-cut pear cuboids were measured after treatment and at 3, 6, 9, 12 and 15 days during storage with a portable PCE-CSM1 reflectance colorimeter (PCE Instruments, UK) calibrated against a white standard. The analysis was performed randomly on three sample pieces from each treatment with three readings on different locations per piece.

The browning index (BI) was used as an indicator of intensity of brown color in fresh-cut fruits and was calculated as described by Kumar et al. [28]:

$$BI = 100\ (x - 0.31)/0.172$$

where $x = (a^* + 1.75L^*)/(5.646L^* + a^* - 3.012b^*)$.

2.7. Firmness

Firmness measurements were carried out right after treatment and at 3, 6, 9, 12 and 15 days during storage. Fruit firmness was measured using a GY-3 fruit penetrometer (Sundoo Instruments, Zhejiang, China) fitted with an 8 mm diameter, round plunger tip. Eight pear cuboids were analyzed for each coating treatment and the mean value was reported in kg/cm^2.

2.8. Titratable Acidity and Soluble Solids Content

Titratable acidity and soluble solids content were measured after treatment and at 3, 6, 9, 12 and 15 days during storage. The titratable acidity was determined in 10 g of homogenate from three pear cuboids, made up to 100 g with distilled water and titrated to pH 8.2 with 0.1 N NaOH solution. Two independent extracts were prepared and each one was titrated in duplicate. The results were expressed as grams of malic acid per 100 g fresh weight. The content of total soluble solids was determined using a digital refractometer (Hanna Instruments, Woonsocket, RI, USA) and the results were expressed as percentages. Six replications were used for each treatment.

2.9. Total Phenolic Content

The methanolic extract of pears was prepared by homogenizing 3 g of fruit flesh in 10 mL of methanol in an ultrasonic bath for 60 min at room temperature. The homogenate was centrifuged at 6000 rpm for 15 min. The supernatants were transferred to vials, stored at −20 °C and later used for total phenolic content and DPPH free-radical-scavenging activity.

Total phenolic content was assessed according to the Folin–Ciocalteu procedure as described by Singleton et al. [29]. Aliquots of extracts (0.1 mL) were mixed with 5 mL of distilled water and 0.5 mL of Folin–Ciocalteu reagent. After 3 min, 1.5 mL of sodium carbonate solution (20% *w*/*v*) were added and the final mixture was made up to a volume of 10 mL with distilled water. The vials were vortexed and placed in a dark place for

30 min at 40 °C, then the absorbance was measured at 765 nm on a Varian Cary 50 UV spectrophotometer (Varian Co., Palo Alto, CA, USA). Results were expressed as milligrams of gallic acid equivalents (GAE) per 100 g fresh weight based on a standard curve of gallic acid (0.05–0.25 mg/mL). Three replicates were carried out for each sample.

2.10. DPPH Free-Radical-Scavenging Activity

The DPPH free-radical-scavenging activity of the extracts was evaluated based on the method described by Oliveira et al. [30]. Aliquots of fruit extracts (50 µL) were mixed with 3 mL of 0.0004% DPPH methanolic solution. After incubation in darkness for 30 min, the absorbance was read at 517 nm using a Varian Cary 50 UV spectrophotometer (Varian Co., USA) against a blank of methanol without the DPPH reagent. DPPH radical-scavenging activity was calculated according to the following formula:

$$\text{DPPH scavenging activity (\%)} = [1 - \text{absorbance of sample/absorbance of blank}] \times 100$$

Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid) was used as a standard reference, and results were expressed as mmol Trolox equivalents (TE) per 100 g fresh weight.

2.11. Sensory Analysis

Control and treated fresh-cut pears were evaluated after treatment, as well as after 6 and 12 days of storage at 8 °C, in terms of appearance, texture, taste, flavor and overall acceptability. A 5-point hedonic scale was used with 1 meaning "dislike extremely" and 5 meaning "like extremely". Samples receiving scores below 3 for any of the evaluated attributes were considered as unacceptable from a sensory point of view [28]. The panel consisted of 12 members selected from staff and master's degree students of the Department of Food Science at the University of Craiova (Craiova, Romania). The samples were served to the panelists on a white plate immediately after removal from cold storage. The sensory analysis was carried out at ambient temperature under white light. The order of the samples was randomized for each panelist. The evaluations were made in triplicate and the average response was calculated for each sensory attribute.

2.12. Statistical Analysis

All experiments in this study were conducted in a randomized design with at least three replicates and results were expressed as means ± standard deviations. Statistical analysis of the influence of different coating formulations on the fresh-cut pear quality was carried out at each sampling interval (0, 3, 6, 9, 12 and 15 days) using Statgraphics Centurion XVI software (StatPoint Technologies, Warrenton, VA, USA). Statistical significance was assessed by one-way analysis of variance. Multiple comparisons among the treatments with significant differences tested in ANOVA were conducted by using the least-significant-difference LSD test and differences at $p < 0.05$ were considered to be significant.

3. Results and Discussion

3.1. Weight Loss

Moisture loss is an indicator of freshness as it is associated with the decrease in turgescence and crispness of the fresh-cut fruit [28]. The main cause of weight loss in fresh-cut fruits during storage is the evaporation of moisture through the surface of the fruit slices [8,31]. Edible coatings are considered to reduce the water loss in fresh-cut fruits due to their behavior as semipermeable barrier against moisture and one of the main quality parameters of the coating formulation is its potential to decrease weight loss during storage [32]. The effect of the application of pectin coating followed by chemical dips on the quality of fresh-cut pears was evaluated. Uncoated pears and pears coated with pectin were used as controls. Figure 1 shows the evolution of the percentage weight loss during 15 days of storage for both control and coated samples.

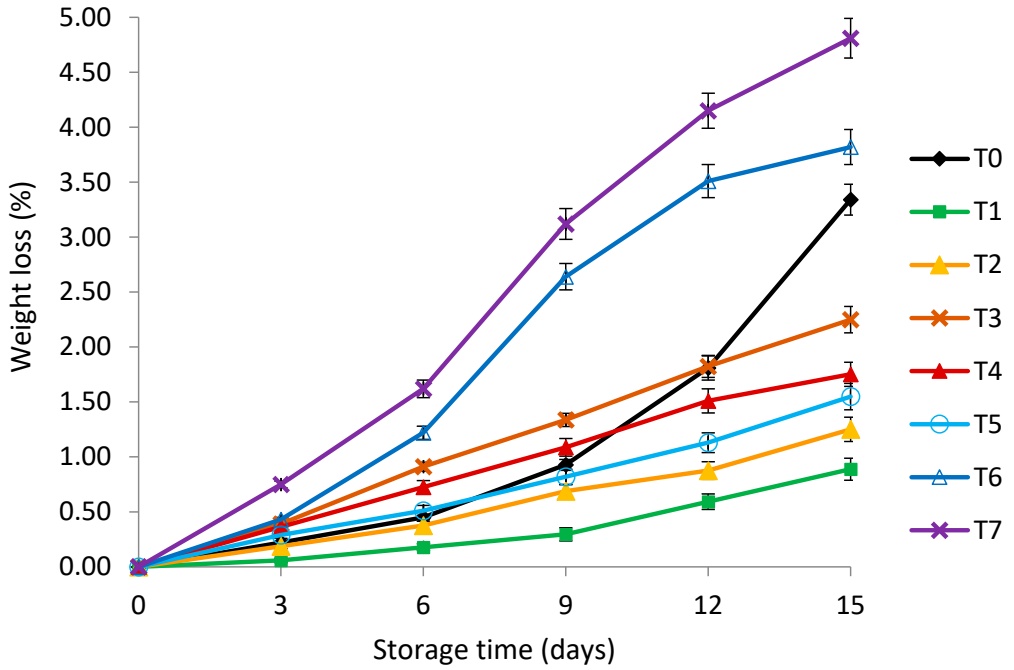


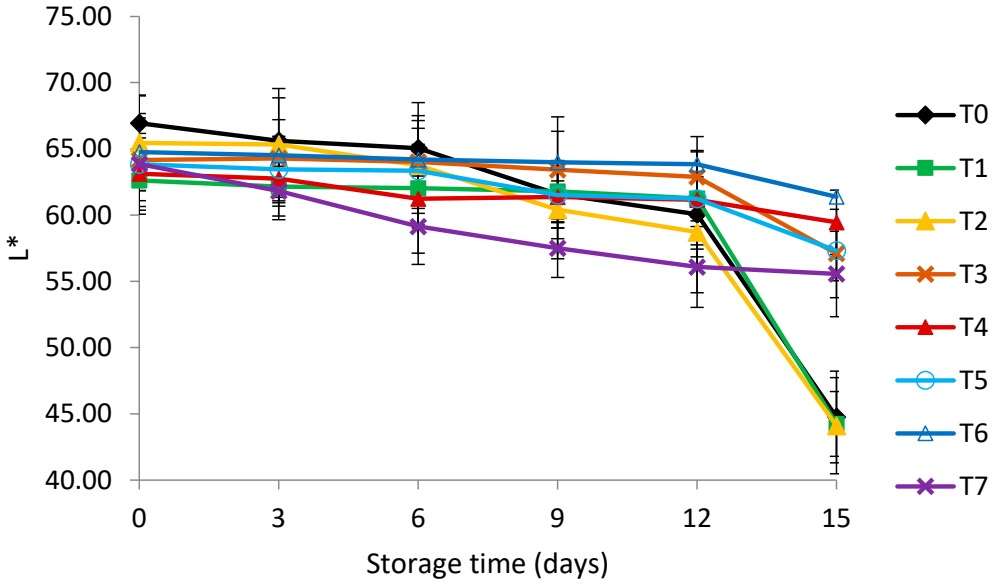
Figure 1. Weight loss of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 9 measurements (3 experiments × 3 replicates). Error bars represent ± SD.

As found in many previous studies on fresh-cut fruits, the weight loss of fresh-cut pear samples increased throughout the refrigerated storage period [28,33]. The weight loss of the pear cuboids found in this study was in the range 0.89–4.81% during the 15 days of storage. The best control of weight loss throughout storage was obtained for the samples coated with pectin alone. After 15 days of storage, control samples lost 3.34% of their weight, while pectin-coated pear cuboids had significantly lower weight loss (0.89%). Dipping in solutions containing chemical preservatives (0.2% PS or 0.2% SB) after coating with pectin contributed to higher weight loss during storage as compared to the samples coated only with pectin, but weight loss values remained lower compared to the uncoated control samples. This could be due to the partial diffusion of pectin from the coating in the solution containing $CaCl_2$ and preservative (0.2% PS or 0.2% SB) during the chemical dip, resulting in the thinning of the edible coating. Kuwar et al. [9] also found that fresh-cut papaya treated with aloe vera gel in combination with a chemical dip containing 2% $CaCl_2$, 1% AA and 8 mM vanillin had higher weight loss during storage than samples coated with aloe vera gel without the chemical dip. The formulations incorporating antibrowning agents (1% *N*-AC or 1% AA + 1% CA) were less effective in controlling weight loss in comparison with the other coated samples. T6 and T7 recorded the highest weight loss throughout storage, significantly higher than the weight loss of the control.

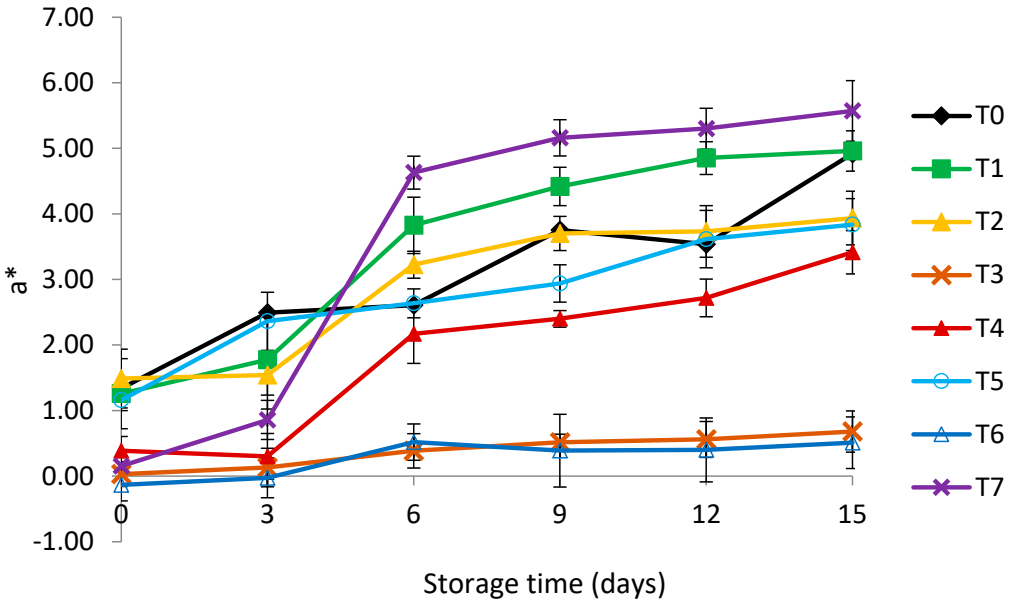
3.2. Color

Color is an important criterion in the quality evaluation of fresh-cut fruits during their shelf life [2]. The effect of pectin coating and pectin coating + chemical dips on the surface-color evolution of pear cuboids was evaluated during the refrigerated storage for 15 days by monitoring the changes in L* (lightness/darkness), a* (redness/greenness), b* (yellowness/blueness) and browning index (BI) (Figure 2). The decrease in lightness after coating has been previously reported in fresh-cut fruits and it was attributed to modifications in the surface reflection properties of the fruit slice that occur as the coating turns opaque after drying [33].

L* values generally decreased during storage as a consequence of the enzymatic and nonenzymatic browning reactions occurring after the tissue damage provoked by peeling and slicing. Moreover, the decrease in L* values has been associated with the loss of water [28]. L* values remained quite stable during the 12 days of storage for the T3 and T6 samples and decreased slightly in the others. In the last three days of storage, the L* values decreased more sharply in samples T3–T7 but dropped dramatically in samples T0, T1 and T2.

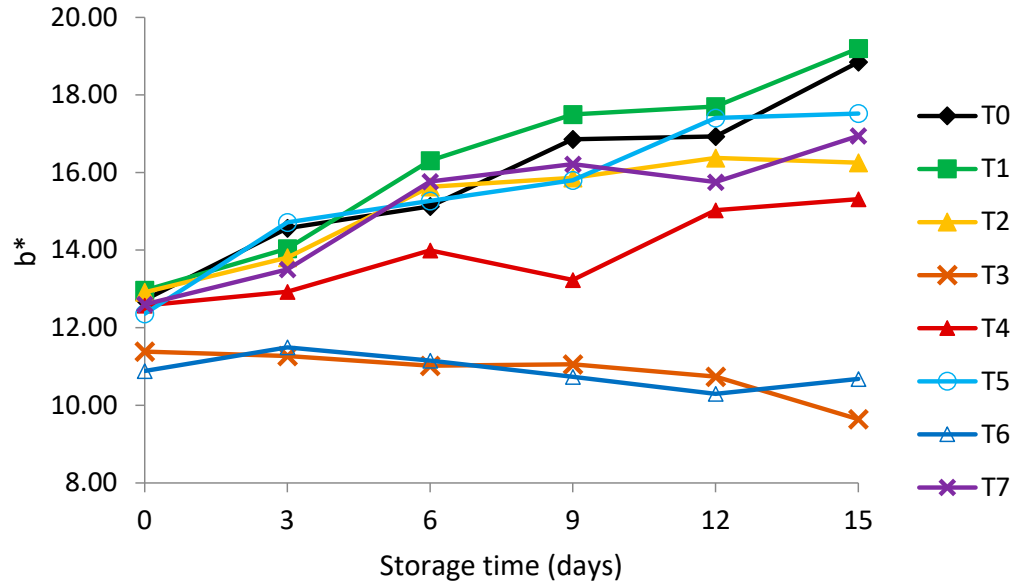


(**a**)



(**b**)

Figure 2. *Cont.*



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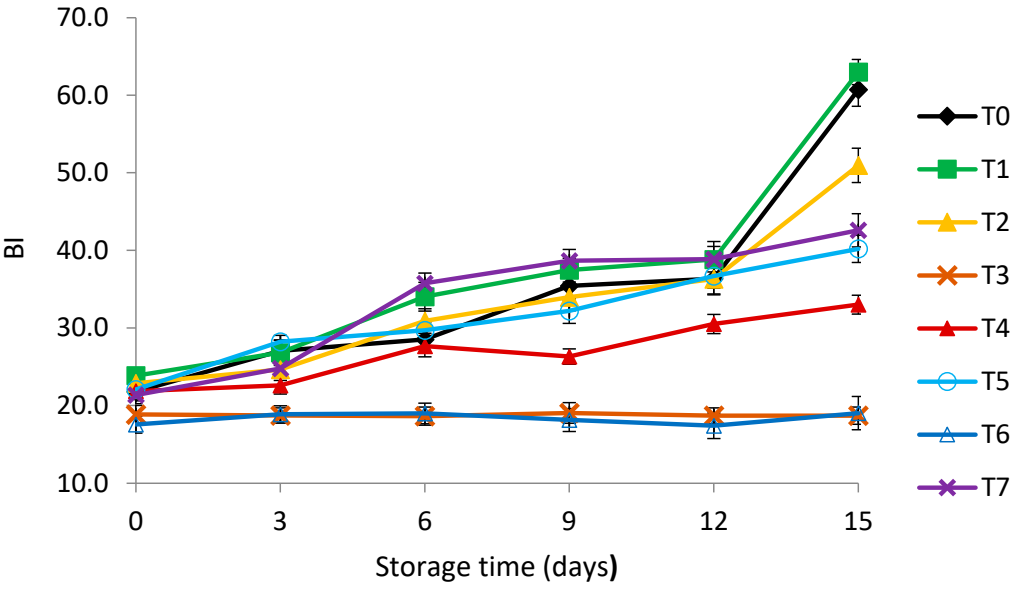

(**d**)

Figure 2. Color values and browning index of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7- 0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. (**a**) Lightness (L*); (**b**) redness (a*); (**c**) yellowness (b*); (**d**) browning index (BI), BI = 100 (x − 0.31)/0.172, x = (a* + 1.75L*)/(5.646L* + a* − 3.012b*). Values are the mean of 27 measurements (3 experiments × 3 replicates × 3 readings/replicate). Error bars represent ± SD.

The results of changes in the a* values of the pear cuboid surfaces during storage (Figure 2b) showed a general increasing trend. The pectin coating combined with dipping in solutions containing 1% *N*-acetyl cysteine (T3 and T6) helped maintain a* values of the pear cuboid surfaces during storage. However, pectin coating alone (T1) and pectin

coating + dipping in solution containing 1% ascorbic acid + 1% citric acid (T4 and T7) resulted in a significant increase in the redness of the pear surfaces after 3 days of storage. After 6 days of storage, the T7 samples presented the highest a* and the lowest L* values. Except T3 and T6 samples, the b* values increased during storage for all samples. The highest increasing trend for b* was observed in the control (T0) and pectin-coated (T1) samples (Figure 2c).

Browning index (BI) is a measure of the intensity of the brown color resulting from the presence of the pigments produced by the browning reactions, and it generally has an upward trend during storage [19]. In the present work, the BI of the control (T0) and pectin-coated (T1) fresh-cut pear cuboids showed a slight increase up to day 12, and then exhibited a rapid rise until the fifteenth day of storage (Figure 2d). Although polysaccharide-based coatings are expected to be good oxygen barriers, the results for L*, a*, b* and BI in this study proved that pectin coating alone did not prevent oxidative browning.

Images of the appearance of fresh-cut pears after 12 days of storage are shown in Figure 3. The pear cuboids dipped in pectin coating solution and crosslinking solution containing antimicrobials and *N*-acetyl-cysteine (T3 and T6 samples) showed similar behaviors throughout the storage duration and maintained the lowest, and quite stable, BI values during the 15 days of storage, indicating the positive effect of these combinations on the control of enzymatic browning. Pectin coating + dipping in 0.2% SB (T5) delayed the increase of BI as compared with the control samples. Pectin coating + 0.2% PS + 1% AA + 1% CA has also been shown to be effective in preventing enzymatic browning. Sanchís et al. [16] developed new edible coatings based on apple pectin and a combination of antioxidants and antimicrobial agents to control enzymatic browning and microbial growth in fresh-cut persimmon. They also found that the addition of 0.2% potassium sorbate helped maintain less browning and suggested a synergic effect of its active form (sorbic acid) with antioxidants.

Figure 3. Appearance of fresh-cut pears after 12 days of storage at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA.

Several previous studies reported that the addition of additives such as antioxidants, acidulants and preservatives to different coatings may reduce browning in fresh-cut fruits [15,28,34]. Guerreiro et al. [32,34] found that ascorbic acid and citric acid were the most efficient antibrowning agents added to pectin coatings for reducing the browning of fresh-cut apples, while Oms-Oliu et al. [12] reported that the incorporation of *N*-acetyl cysteine was effective in preventing fresh-cut pears from browning for 2 weeks without

affecting their firmness. Robles-Sánchez et al. [35] also reported that the treatment of mango cubes with an edible alginate coating containing ascorbic and citric acids as antibrowning agents maintained higher color values (L* and °Hue) compared to mango cubes coated only with alginate or control.

3.3. Firmness

Figure 4 shows that the firmness of the untreated pear cuboids decreased significantly after 15 days of storage. The decrease in firmness is generally caused by metabolic processes such as the hydrolysis of starch and the degradation of pectin in the fruit cell wall as a result of pectic-acid hydrolysis [7]. Another factor involved in the changes in firmness during storage is moisture loss, which may lead simultaneously to a hardening and a decrease in the turgor and crispness of fresh-cut fruits [28,31,36]. Fresh-cut pear samples coated with pectin and dipped in 2% $CaCl_2$ maintained higher firmness as compared with control samples, a fact that may be attributed to the supplementary effect of the edible coating on the surface of the fruit. No significant differences were found between the firmness of the treated samples in the first 9 days of storage. However, at the end of the storage period, the loss of firmness in the control samples was around 53%, whereas the firmness of dipped + coated samples decreased by 2–31% (Figure 4). In the pectin-coated samples dipped in a solution containing 1% $CaCl_2$, the firmness increased initially during the first 6 days of storage, then remained stable or slightly decreased through the following 6 days of storage and declined sharply afterwards.

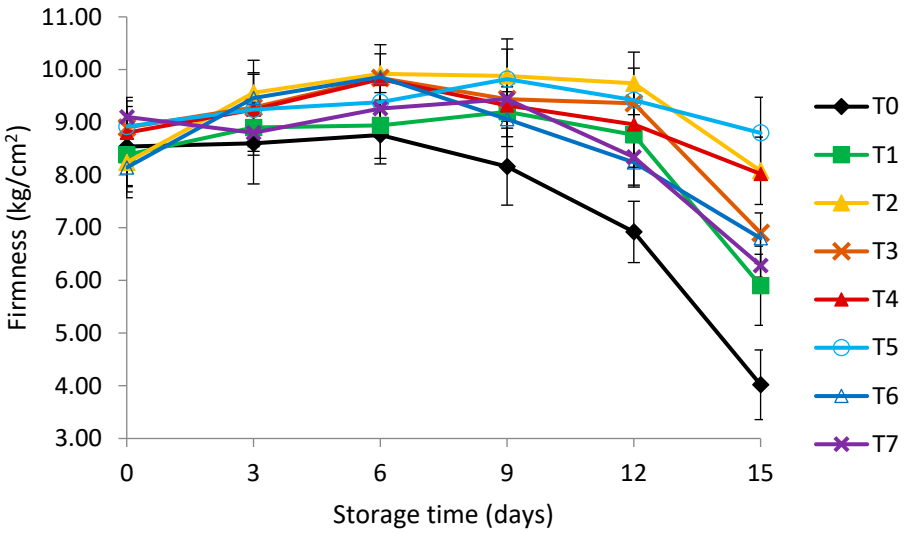

Figure 4. Firmness of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 24 measurements (3 experiments × 8 replicates). Error bars represent ± SD.

The increase in firmness after dipping in $CaCl_2$ solution is in accordance with previous studies and has been attributed to the calcium pectate formed from calcium ions and pectic acid in the cell wall, which strengthens the molecular binding between cell wall constituents [28,37]. The most effective treatments in controlling firmness were those with pectin coating + 2% $CaCl_2$ + 0.2% SB (T5) or pectin coating + 2% $CaCl_2$ + 0.2% PS (T2), but the treatments with the addition of antioxidants also led to good results regarding the changes in firmness. Bico et al. [15] found that the combined effect of alginate or carrageenan coating and chemical dip in 1% $CaCl_2$, 0.5% ascorbic acid and 0.75% cysteine reduced the softening rate in fresh-cut banana compared to the control (untreated) and coating-only samples, while Guerreiro et al. [32] reported the positive effect of coatings incorporating antibrowning agent in the retention of fresh-cut apple firmness. Saba and

Sogvar [38] also found that the treatment of apple slices with carboxy methylcellulose containing $CaCl_2$ and ascorbic acid significantly retained the firmness of fresh-cut apples.

3.4. Titratable Acidity (TA) and Total Soluble Solids (TSS)

The changes in TA and TSS during the storage of fresh-cut pear cuboids are shown in Figures 5 and 6, respectively. At day 0 of the storage period, the higher values of TA found in the T4 and T7 samples as compared to control and other treated samples might be due to the incorporation of AA and CA as components of the chemical dip.

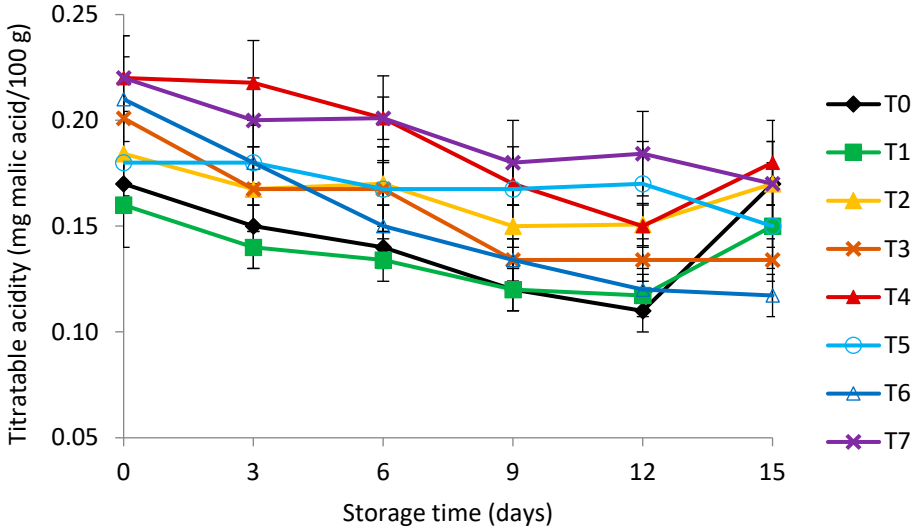

Figure 5. Titratable acidity of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 6 measurements (3 experiments × 2 replicates). Error bars represent ± SD.

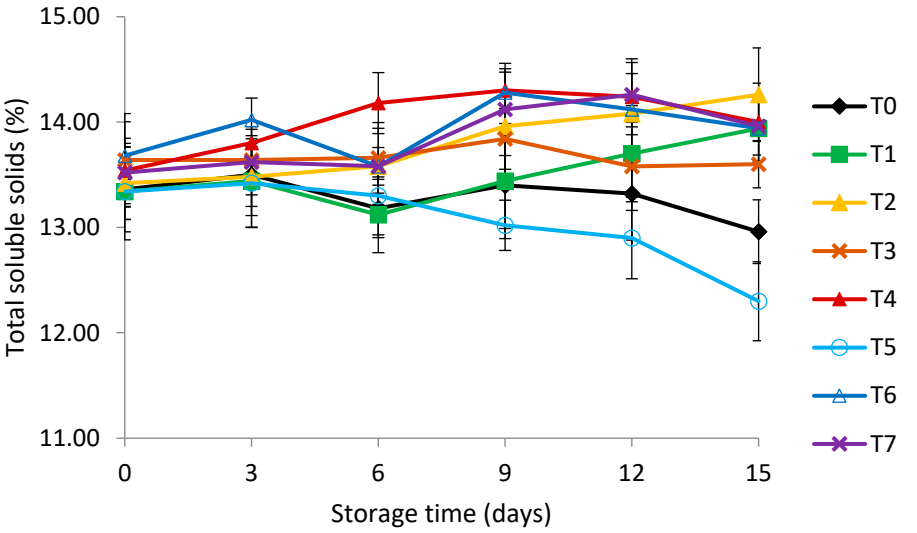

Figure 6. Total soluble solids of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 18 measurements (3 experiments × 6 replicates). Error bars represent ± SD.

The titratable acidity generally decreased during first 12 days of storage in all samples, probably as a result of the conversion of organic acids into sugars and their use as substrates in respiratory metabolism. A similar trend for titratable acidity was also reported for fresh-cut apples [39], nectarines [33,40], peaches [41], pineapple [9], oranges [8] and kiwifruit [42]. It was interesting to note that after 12 days of storage, samples dipped in 0.2% sodium benzoate (T5–T7) did not show an increase in titratable acidity, in contrast to the other samples (T0–T4), where titratable acidity increased, probably as a result of the onset of fermentation occurring in these samples. Titratable acidity was not significantly affected by pectin-coating treatment alone during refrigerated storage ($p > 0.05$).

The pectin-coated samples (T1) and those coated with pectin and dipped in 0.2% preservative (T2 and T5) showed the lowest decrease in titratable acidity in the 12 days of storage. This may indicate that these treatments provided a gas barrier on the fruit surface or decreased surface-gas permeability, leading to a delay in the respiration rate. The addition of citric and ascorbic acids to the chemical dip (T4 and T7) resulted in a significantly higher ($p < 0.05$) titratable acidity of the pear samples throughout storage, which probably contributed, together with the preservatives, to the delay of the fermentation processes in the fruit.

The mean total soluble solid content of the pear cuboids before the coating treatments was 13.5%. At the end of the storage period, most of the treated samples showed significantly higher TSS values, in good agreement with the results reported by Chiabrando and Giacalone [33] for fresh-cut nectarines or by Duan et al. [31] for fresh blueberries. Control (T0) and pectin-coated samples dipped in 0.2% SB (T5) showed the lowest values (12.96% and 12.3%, respectively). TSS slightly increased in the first 3 days in most samples and then tended to decrease until day 6, increasing, however, with longer storage. Control samples showed a decrease in TSS content during the last 3 days of storage. The increase in TSS has been attributed to the acid metabolism that continues during post-harvest storage by converting starch and acid into sugar, while the decrease in TSS after 12 days of storage is the result of the utilization of sugars in the metabolic and degradation processes occurring in the fresh-cut fruits. However, in the last 3 days of storage, TSS was maintained or even slightly increased in most of the pectin-coated samples, both for those coated with pectin alone or combined with chemical dipping. This could be explained by the fact that these edible coatings may slow down the degradative biochemical changes as compared with the uncoated samples. Similar results were previously reported by Benítez et al. [42] and Passafiume et al. [2] for fresh-cut kiwifruit coated with aloe vera gel and or by Kuwar et al. [19] for fresh-cut papaya samples coated with aloe vera gel and honey alone, or for coated papaya additionally pretreated with chemical dip.

3.5. Total Phenolic Content

The total phenolic content increased initially but remained relatively stable or slightly decreased after 3 days of storage, after which it increased again in all samples up to 12 days of storage (Figure 7). A similar variation was found by Oms-Oliu et al. [43] for fresh-cut melon uncoated and coated with alginate, gellan and pectin; they reported that the initial phenolic content was maintained or slightly decreased during the first week of storage but then increased in the second week. No significant differences were determined between the total phenolic content of the T0, T1, T2 and T5 samples during 12 days of storage, showing that edible pectin coatings alone or in combination with antimicrobials did not affect the total phenol content.

However, in the first 12 days of storage, the pear cuboids treated with the edible pectin coating and anti-browning agents (T3, T4, T6 and T7) showed significantly higher total phenolic content as compared with the other samples. These results are in agreement with previous reports, where treatment with edible coatings in combination with dipping in solutions of antibrowning agents was more effective in maintaining total phenolic content during storage for fresh-cut apples [38], mangoes [35] and papaya [19] as compared to treatments with coating alone. Gonzalez-Aguilar et al. [44] also found that antibrown-

ing treatments reduced the loss of phenol content in fresh-cut pineapple. Antibrowning agents protect phenolic compounds by preventing polyphenol oxidase–catalyzed reactions. Moreover, a higher phenolic content in fresh-cut fruits is correlated with a higher ability to scavenge free radicals, thus maintaining quality by preventing oxidative reactions during extended storage. After 12 days of storage, the maximum total phenolic content was found in pectin-coated samples dipped in 0.2% SB + 1% *N*-AC (T6).

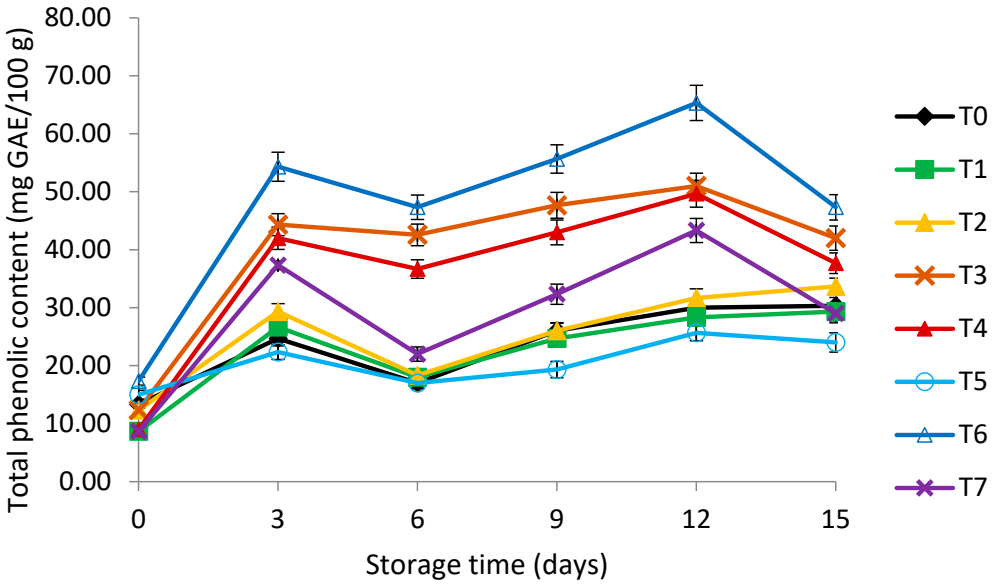

Figure 7. Total phenolic content of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 9 measurements (3 experiments × 3 replicates). Error bars represent ± SD.

3.6. DPPH Radical-Scavenging Activity

Figure 8 shows the DPPH radical-scavenging activity of fresh-cut pears as affected by pectin coating and chemical dips. Neither pectin coating alone nor in combination with preservatives (T1, T2 and T5) significantly influenced the antioxidant activity of fresh-cut pears.

The DPPH antioxidant activity of the treatments containing *N*-acetyl-cysteine (T3 and T6) was significantly higher than that of the other samples immediately after the coating treatment (Figure 8). Oms-Oliu et al. [12] also reported that a pectin coating containing *N*-acetyl cysteine considerably increased the antioxidant activity of fresh-cut pears. In the first days of storage, the antioxidant activity of these samples exhibited a decreasing trend, then there was an increase and finally a decrease. These increases recorded between the sixth and twelfth days of storage may be attributed to the increase in the total phenolic content recorded during this period and caused by the induction of phenylpropanoid metabolism [43,45]. The presence of *N*-acetyl-cysteine as an antioxidant could be behind this behavior, acting as a protective agent against oxidative stress sources and, at the same time, contributing to the activation of phenolic compound production by the fruit tissues. In the last 3 days of storage, the antioxidant activity decreased in these samples, in correlation with the decrease of the phenolic content registered in this period.

The combination of pectin coating with a dipping treatment containing ascorbic acid (T4 and T7) resulted into a consistent initial increase of the DPPH antioxidant activity. These results are supported by previous studies reporting that ascorbic acid included in a dipping treatment significantly increased the antioxidant capacity of fresh-cut fruits [35,45]. However, the antioxidant activity in these samples was significantly lower ($p < 0.05$) than that of the pectin-coated samples dipped in 1% *N*-acetyl cysteine (T3 and T6). In the first 6

days of refrigerated storage, the DPPH radical-scavenging activity decreased sharply in samples dipped in 1% AA + 1% CA (T4 and T7). After this time, the antioxidant activity remained stable and no significant differences were found between the T4 and T7 samples and the others, except those dipped in 1% *N*-acetyl cysteine (T0, T1, T2 and T5). Duan et al. [31] also reported decreases in the antioxidant activity in the first 6 days of storage for coated and uncoated fresh blueberries followed by a stable trend in the next 6 days. The decrease in antioxidant activity values may be attributed to the oxidation of antioxidant compounds such as ascorbic acid and polyphenols, which may be easily degraded in the presence of oxygen by enzyme-mediated reactions [27].

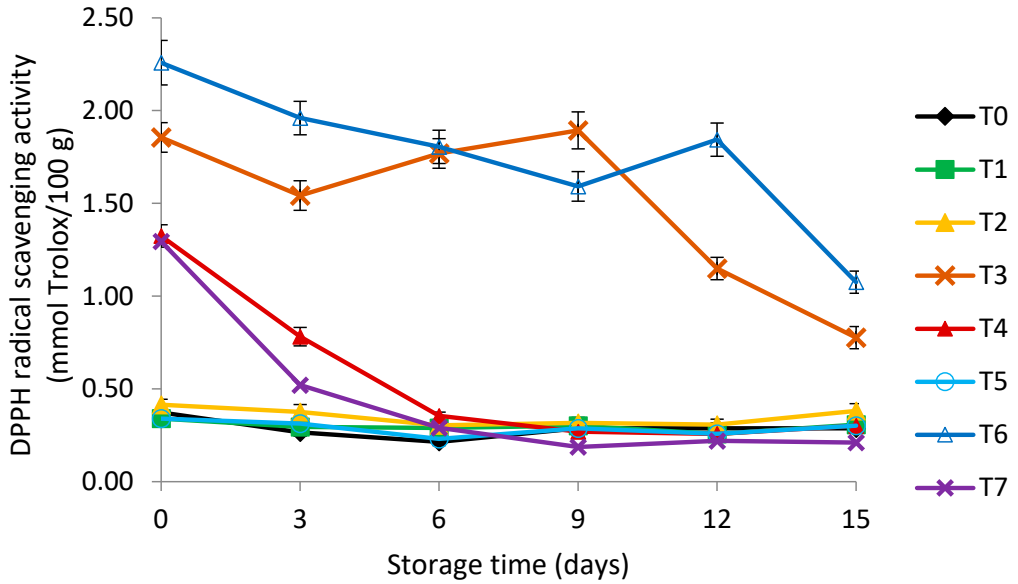

Figure 8. DPPH radical-scavenging activity of control and treated fresh-cut pears stored for 15 days at 8 ± 1 °C. T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values are the mean of 9 measurements (3 experiments × 3 replicates). Error bars represent ± SD.

3.7. Sensory Analysis

Table 2 shows the changes in appearance, texture, taste, flavor and overall acceptability scores for control and treated fresh-cut pears stored for 12 days at 8 °C. The scores for appearance decreased during storage for all samples as a result of the darkening at the surface and of the loss of freshness and moisture [9]. An obvious reduction in the sensory scores during storage was also reported in pectin-coated apples by Moreira et al. [27] and by Sanchís et al. [16] for fresh-cut persimmon. Significant differences ($p < 0.05$) were detected in all sensory attributes between samples, even just after the treatments.

Dipping in solutions with antibrowning agents combined with edible pectin coating determined an increase in the sensory scores. The samples coated with pectin and dipped in 1% *N*-acetyl-cysteine (T3 and T6) received the highest scores for all sensory attributes, both after 6 and 12 days of storage (Table 1). In addition, the T3 and T6 samples were the only ones with scores exceeding the limit of acceptability (3.0) for all sensory parameters after 12 days of storage. When using 1% AA + 1% CA as antibrowning agent (T4 and T7), the appearance was lower than when using 1% *N*-acetyl-cysteine, both in combination with potassium sorbate and with sodium benzoate (T3 and T6). Scores for fresh-cut pears coated with pectin and dipped in 0.2% PS (T2) were not significantly different from those for samples coated with pectin alone (T1) for all sensory parameters evaluated. However, the same cannot be said for samples coated with pectin and dipped in 0.2% SB (T5), which received lower scores for taste and flavor. Sanchís et al. [16] noticed that adding

antimicrobials and antioxidants to pectin coatings conferred slight acidity and "off-flavor" to fresh-cut persimmon samples.

Table 2. Scores for sensory analysis of control and treated fresh-cut pears at 0, 6 and 12 days of storage at 8 °C *.

Treatment	Storage Time (Days)	Appearance	Texture	Taste	Flavor	Overall Acceptability
T0	0	4.25 ± 0.45^{ab}	4.50 ± 0.52^{abc}	4.83 ± 0.39^{e}	4.67 ± 0.49^{d}	4.58 ± 0.51^{ab}
	6	3.75 ± 0.45^{c}	3.08 ± 0.29^{a}	3.83 ± 0.39^{d}	3.42 ± 0.51^{ab}	3.42 ± 0.51^{b}
	12	2.42 ± 0.51^{b}	2.17 ± 0.39^{a}	2.75 ± 0.45^{ab}	2.83 ± 0.49^{ab}	2.17 ± 0.39^{b}
T1	0	4.08 ± 0.29^{a}	4.25 ± 0.45^{a}	4.50 ± 0.52^{de}	4.42 ± 0.51^{bcd}	4.50 ± 0.52^{ab}
	6	3.58 ± 0.51^{bc}	3.67 ± 0.49^{bc}	3.67 ± 0.49^{bcd}	3.67 ± 0.49^{ab}	3.25 ± 0.45^{b}
	12	2.58 ± 0.39^{b}	2.75 ± 0.45^{cd}	2.83 ± 0.39^{abc}	2.75 ± 0.51^{ab}	2.33 ± 0.49^{b}
T2	0	4.33 ± 0.49^{ab}	4.42 ± 0.51^{abc}	4.33 ± 0.49^{cd}	4.33 ± 0.49^{bcd}	4.33 ± 0.49^{ab}
	6	3.50 ± 0.52^{bc}	3.83 ± 0.39^{c}	3.42 ± 0.51^{abc}	3.58 ± 0.51^{ab}	3.17 ± 0.39^{b}
	12	2.67 ± 0.49^{b}	2.83 ± 0.39^{d}	2.67 ± 0.49^{a}	2.92 ± 0.51^{ab}	2.42 ± 0.51^{b}
T3	0	4.83 ± 0.39^{c}	4.67 ± 0.49^{bc}	4.17 ± 0.39^{bcd}	4.50 ± 0.52^{cd}	4.67 ± 0.49^{b}
	6	4.58 ± 0.51^{d}	4.42 ± 0.51^{d}	3.92 ± 0.67^{d}	3.75 ± 0.45^{bc}	4.25 ± 0.62^{c}
	12	3.92 ± 0.29^{c}	3.58 ± 0.51^{e}	3.17 ± 0.39^{d}	3.08 ± 0.51^{b}	3.42 ± 0.51^{c}
T4	0	4.58 ± 0.51^{bc}	4.58 ± 0.51^{abc}	4.08 ± 0.67^{bc}	4.58 ± 0.51^{d}	4.58 ± 0.51^{ab}
	6	3.33 ± 0.49^{b}	3.83 ± 0.39^{c}	3.42 ± 0.51^{abc}	4.08 ± 0.29^{c}	3.08 ± 0.29^{b}
	12	2.33 ± 0.49^{b}	2.67 ± 0.49^{bcd}	2.83 ± 0.39^{abc}	2.87 ± 0.52^{ab}	2.42 ± 0.51^{b}
T5	0	4.08 ± 0.29^{a}	4.33 ± 0.49^{ab}	3.92 ± 0.29^{ab}	3.92 ± 0.29^{a}	4.25 ± 0.45^{a}
	6	3.42 ± 0.51^{bc}	3.50 ± 0.52^{bc}	3.33 ± 0.49^{ab}	3.33 ± 0.49^{a}	3.17 ± 0.39^{b}
	12	2.42 ± 0.51^{b}	2.42 ± 0.51^{abc}	2.67 ± 0.49^{a}	2.67 ± 0.39^{a}	2.33 ± 0.49^{b}
T6	0	4.75 ± 0.45^{c}	4.75 ± 0.45^{c}	4.08 ± 0.67^{bc}	4.17 ± 0.58^{abc}	4.58 ± 0.51^{ab}
	6	4.67 ± 0.49^{d}	4.33 ± 0.49^{d}	3.75 ± 0.45^{cd}	3.50 ± 0.52^{ab}	4.17 ± 0.58^{c}
	12	3.67 ± 0.49^{c}	3.33 ± 0.49^{e}	3.08 ± 0.51^{cd}	3.00 ± 0.52^{ab}	3.33 ± 0.49^{c}
T7	0	4.33 ± 0.49^{ab}	4.42 ± 0.51^{abc}	3.67 ± 0.49^{a}	4.08 ± 0.29^{ab}	4.33 ± 0.49^{ab}
	6	2.83 ± 0.39^{a}	3.42 ± 0.51^{ab}	3.25 ± 0.45^{a}	3.42 ± 0.51^{ab}	2.58 ± 0.51^{a}
	12	1.83 ± 0.39^{a}	2.33 ± 0.49^{ab}	2.58 ± 0.51^{a}	2.75 ± 0.45^{ab}	1.67 ± 0.49^{a}

* T0—Control; T1—0.2% PE + 1% $CaCl_2$; T2—0.2% PE + 1% $CaCl_2$ + 0.2% PS; T3—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% *N*-AC; T4—0.2% PE + 1% $CaCl_2$ + 0.2% PS + 1% CA + 1% AA; T5—0.2% PE + 1% $CaCl_2$ + 0.2% SB; T6—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% *N*-AC; T7—0.2% PE + 1% $CaCl_2$ + 0.2% SB + 1% CA + 1% AA. Values for the same storage time followed by different lowercase letter are significantly different at $p < 0.05$ using LSD test. Data are the means ± standard deviation of three independent experiments.

The control samples initially obtained the highest scores for taste and flavor, but after 12 days of storage, the scores for all attributes of the control samples fell below the threshold of acceptability (3.0) as a result of moisture loss and degradative processes such as phenol oxidation and microbial spoilage.

The most prominent decreases in the sensory scores after storage were noticed in the pectin-coated samples dipped in 0.2% SB + 1% AA + 1% CA (T7), in agreement with the highest weight loss and browning index recorded for these samples. This evolution could be related to post-harvest physiological changes induced by the high titratable acidity and by the trapping of volatile compounds in the internal atmosphere of the cut fruit.

4. Conclusions

Pectin coating followed by dipping in solutions containing 2% $CaCl_2$ was effective in controlling the weight loss and firmness of fresh-cut pears when compared to uncoated samples. The incorporation of 1% *N*-AC or 1% AA + 1% CA as antibrowning agents in the formulation of the chemical dip following pectin coating contributed to the protection of the phenolic content and to the enhancement of the antioxidant activity of fresh-cut pears during storage. Pectin coating combined with chemical dip containing 0.2% SB or 0.2% PS and 1% *N*-AC were the most effective treatments in preserving color and reducing browning index in fresh-cut pears over 12 storage days at 8 °C. These treatments allowed fresh-cut pears to reach prolonged storage periods with sensory scores above the sensory acceptability threshold (3.0) for all the attributes evaluated. The results of this work demonstrate that pectin-based edible coatings followed by chemical dip with the incorporation of 0.2% SB or 0.2% PS as antimicrobials and 1% *N*-AC as an antibrowning agent provide the potential to extend the shelf life of fresh-cut pears.

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