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The effects of chitosan biofilm in combination with vinegar, sodium alginate, and carboxymethyl cellulose on the transformation of biochemical and physiological indexes of pears during storage

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Abstract

Pears have a characteristically delicious taste and are rich in nutritional value, especially sugar, vitamin C, vitamin A, and minerals. This study aimed to determine the amount of chitosan-derived probiotics that can be combined with edible vinegar and sodium alginate carboxymethyl cellulose to create a pear preservation process. Formulated chitosan in combination with table vinegar, sodium alginate, and carboxymethyl cellulose at a rate of 160 g chitosan/160 mL table vinegar/7,000 mL water, 4 g sodium alginate, and 0.5 g carboxymethyl cellulose to preserve 180 kg of pears. In 5 preservation test formulations, it was determined that the content of probiotics at 1.0% is suitable for preserving pears. When stored in the content of this probiotic, it has limited the transformation of biochemical and physicochemical indexes. Especially by the fourth week of storage, pears still retained their characteristic values; the natural loss rate was only 5.23% and the rot rate was 5.07%, both lower than when stored in other biological products. In comparison to fresh pears before being put into storage, the change in biochemical and physicochemical indexes of preserved pears with a biological content of 1.0% is only a little.

1. Introduction

The scientific name of the pear tree is *Pyrus pyrifolia Nakai*, which is widely grown in Vietnam, especially in Cao Bang, Bac Kan, Lang Son, Lao Cai, and Lai Chau provinces. In Cao Bang province, pear trees are grown in Thach An, Tra Linh, Bao Lac, Nguyen Binh, and Ha Quang districts, with a total area of 131.81 ha and an average yield of 3.18 tons per ha. Particularly, Thach An district now has an area of pear trees totaling 90 hectares, concentrated in Le Lai, Le Loi, Duc Long, Van Trinh, and Dong Khe communes. The pear is spherical and evenly rounded; the ripe rind has dark brown spots on a yellowish brown background; the height of the fruit is 65 –72 mm; and the average weight of the fruit is 400–450 g (Francisco *et al.*, 2017; Hussain *et al.*, 2021). Pear has good effects on health, bones, and joints; improves memory; supports digestion; protects the heart; supports weight loss; prevents cancer; and improves vision (Francisco *et al.*, 2017; Hussain *et al.*, 2021). After harvesting, pears continue to undergo biochemical, physicochemical, physiological, and respiratory changes. Unlike fruits on trees, biochemical and physicochemical processes occur for pears during storage, mainly breaking down organic compounds to provide energy to maintain cell life (Shehzad *et al.*, 2015). When the pear is ripe, if it is not harvested in time but still left on the tree, it will cause the fruit to be dehydrated, porous, rotten, and reduce the quality of the fruit; at the same time, when leaving the pear on the tree, the tree must provide nutrition to feed the fruit, thereby affecting the yield of the following crop (Shehzad *et al.*, 2015; Francisco *et al.*, 2017). Currently, there are a number of studies on pear preservation in the world. For example, the author Nguyen L.L.P. and his colleagues have applied ethylene to ripen pears treated with 1-MCP after refrigeration (Gamrasni *et al.*, 2010; Nguyen *et al.*, 2022), and Lysiak *et al.* (2021) have studied the effect of storage conditions on the shelf life and antioxidant capacity of pears. In addition, there are some studies using chitosan membranes to preserve vegetables; for

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example, Kumarihami *et al.* (2022) applied chitosan as an edible coating to improve the preservation and quality of kiwi, and the results show that chitosan has been effective in preserving kiwifruit. Currently, pears are mainly preserved cold and used for fresh eating. The research to preserve pears by biotechnology now has very few published studies. Therefore, the application of chitosan biofilm combined with vinegar to preserve pears is essential, contributing to prolonging the shelf life of pears, saving costs, and being environmentally friendly. Chitosan is an animal polymer extracted from shrimp, crab, and crab shells. Chitosan is of natural origin, biodegradable, non-toxic, easy to form into films, and safe for human consumption in food, pharmaceuticals, and cosmetics. Chitosan has a variety of biological effects: the ability to absorb water and moisturize, antifungal properties, and strong antibacterial properties with many different types. Vinegar has been produced for a long time and can be produced on an industrial scale or by fermentation. Edible vinegar is used a lot in food processing and preservation; it has a strong antiseptic effect, inhibiting the growth of mold. After combining these two components together with water, it will form a yellow, viscous liquid that will form a film to preserve the pear. This membrane has a characteristic yellow color and a homogeneous viscous liquid form, ensuring food safety and hygiene, being easy to decompose, and not polluting the environment (Loi and Tung, 2019). In this study, the content of chitosan probiotics was combined with edible vinegar, sodium alginate, and carboxymethyl cellulose at the rate of 160 g of chitosan/160 mL of edible vinegar/7,000 mL of water, 4 g of sodium alginate, and 0.5 g of carboxymethyl cellulose to preserve 180 kg of pears grown in Thach An district, Cao Bang province. Therefore, it is necessary to determine the effect of chitosan biofilm in combination with vinegar, sodium alginate, and carboxymethyl cellulose on the transformation of biochemical and physicochemical indexes of pears during storage, which is of high scientific and practical significance. Since then, it has served as the basis for building a technological process to preserve pears with this biofilm.

2. Materials and methods

2.1 Materials

Raw materials: pears harvested at 200–210 days of age since fruiting at pear farms in Duc Long commune, Le Loi commune, Thach An district, and Cao Bang province. The pear is spherical and evenly rounded; the ripe skin has dark brown spots on a yellowish brown background; the height of the fruit is 65–72 mm; and the average weight of the fruit is 400–450 g.

Film-forming materials: Membrane materials including chitosan, vinegar, clean water, sodium alginate, and carboxymethyl cellulose ensure quality standards. Chitosan, vinegar, and clean water originate in Vietnam; sodium alginate originates in the UK; and carboxymethyl cellulose originates in China.

Chemicals used in analysis: Chemicals used in the study include: Na_2SO_3, phenolphthalein 1%, NaOH 10% solution, HCl acid 15%, potassium ferricide 1%, KOH 2.5N, methylene blue, iodine 0.1N solution, starch 1% indicator, tryptone, high yeast concentration, etc. These chemicals ensure quality and originate in Vietnam.

2.2 Sampling method

The pears are sampled according to Vietnamese standard TCVN 9017:2011. Fresh Fruit Sampling Method in the Production Garden (Vietnamese National Standards, 2011). At the time of sampling pears in the afternoon, the weather is dry; there is no rain and no fog. The pear must meet quality standards, and be free from pests, mold, injury, and black spots. The pear is then packed in a foam container, perforated, and transported back to the laboratory for preservation studies. The time it takes to transport samples from the sampling site to the storage laboratory is about 6–6.5 hrs.

2.3 Biological products production

The composition of the preservation film includes 160 g of chitosan, 160 mL of edible vinegar, 7,000 mL of water, 4 g of sodium alginate, and 0.5 g of carboxymethyl cellulose to preserve 180 kg of pears. The process of making probiotics is carried out as follows: While stirring, dissolve the chitosan in the vinegar, then slowly pour in the water and stir well, allowing it to stabilize at room temperature for 72 hrs. 4 g of sodium alginate and 0.5 g of carboxymethyl cellulose were dissolved into 200 mL of water at 60°C using a magnetic stirrer until the materials were completely dissolved, then allowed to cool. Next, add 4 mL of glycerol, 0.4 mL of tween-80, and 2 mL of 2% $CaCl_2$ solution to the mixture of sodium alginate and carboxymethyl cellulose, then stir until the solution is homogeneous (about 60 mins) and mix well into the solution (chitosan, vinegar, and water) to form a homogeneous mixture (Loi and Tung, 2019).

2.4 Experiment arrangement

The experiment was arranged according to 5 formulas, repeated 3 times and shown in Table 1. The pear is dipped directly into the biological preparation; each batch's dipping time is 2 mins; the pear is then picked up and placed in a plastic basket to dry the film before being stored at natural temperature conditions. Dry, cool, avoid direct sunlight and rain, and these pears are stored at natural temperature conditions. Once a week, samples are analyzed using analytical indexes such as the variation in soluble dry matter content, vitamin C, total sugar content, total protein content, respiratory intensity variation, color variation, natural mass loss, and rotten pear rate (Loi and Tung, 2019).

Table 1. Experimental recipes for pears storage.

TT	Experimental elements	Experimental formulas				
		CT-1	CT-2	CT-3	CT-4	CT-5
1	Weight of pear (kg)	50	50	50	50	50
2	Film composition content (%)	0	0.5	1	1.5	2

2.5 Analytical methods

2.5.1 Dissolved dry matter content

The dissolved dry matter content was determined by the Japanese ATAGO N-1α photometer, which is measured in Brix (°Bx) at 20°C. When light passes through a solution of different soluble dry matter, the light is refracted at different refractive angles, from which the dry matter concentration of the analyte can be deduced (Loi and Tung, 2019).

2.5.2 Vitamin C content

The vitamin C content of pears is determined according to Vietnamese Standard TCVN 6427-2:1998 (ISO 6557/2: 1984). This method was carried out according to the principle of extracting ascorbic acid from pears with a solution of oxalic acid. The titration was 2.6 dichlorophenolindophenol until the solution was pale pink (Vietnamese National Standards, 1998).

2.5.3 Total sugar content

The total sugar content of pears is determined according to Vietnamese Standard TCVN 4594-88:1988. This method is carried out according to the principle of extracting the total sugar from the sample with hot water, using hydrolyzed hydrochloric acid to produce glucose sugar, which is determined through reactions with phelating solution, iron 3 sulfate, and potassium pemanganate (Vietnamese National standards, 1988).

2.5.4 Total protein content

The total protein content of pears is determined according to Vietnamese Standard TCVN 9936: 2013. This method is carried out in principle by decomposing the organic matter with sulfuric acid in the presence of a catalyst, alkalizing the reaction products, distilling the released ammonium, and retracting it into a solution of boric acid, which is then titrated with a standard sulfuric acid solution (Vietnamese National standards, 2013).

2.5.5 Respiratory intensity

The ICA15 DUAL Analyser Respiratory Intensity Meter measures the pear's respiratory intensity. The respiratory intensity of the pear was determined through the analyses by measuring the amount of CO_2 produced by the respiratory strength meter. The respiratory intensity of a pear is calculated as the amount of CO_2 produced per kilogram of product in one unit of time (Loi and Tung, 2019).

The respiratory rate of pear is calculated according to the following formula:

$$X = \frac{A.V}{1000.m.t.100}$$

Where X = respiratory intensity (mL CO_2/kg/h), A = measured CO_2 concentration (%), m = sample weight (kg), t = time (hours), 1000 = conversion factor from g to kg, and V = volume of free air in the respirometer (mL) Loi and Tung, 2019).

2.5.6 Color variation

The color variation of pear shells through each stage was determined with the Nippon Denshoku NR 300 handheld colorimeter (Japan), based on the principle of light analysis. For each sample, the machine will give the measurement results showing the indexes L, a, b. The color variation of the fruit is determined by the formula: $\Delta E = [(L_i-L_o)^2+(a_i-a_o)^2+(b_i-b_o)^2]^{1/2}$. In which: L_i, a_i, b_i: Color measurement results at the i th analysis, L_o, a_o, b_o: Color measurement results of input materials (Loi and Tung, 2019).

2.5.7 Natural mass loss

The natural mass loss of pears was determined by weighing each fruit in each recipe before storage and after each follow-up. The natural mass loss was calculated by the formula: $X = (M_1-M_2)/M_1$. In which, X: The rate of natural mass loss at each follow-up (%); M_1: The volume of preserved fruits (g); M_2: The volume of fruits at follow-ups (g) (Loi and Tung, 2019).

2.5.8 Rot rate

The rotten rate of pears is determined by the calculation method as follows (Loi and Tung, 2019):

$$\text{Rotten rate (\%)} = \frac{A}{B} \times 100$$

Where: A is the number of rotten fruits, B is the number of tracking fruits.

2.6 Statistical analysis

One-way analysis of variance was performed with Minitab software version 16. The least significant

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differences in mean comparisons were calculated at $p < 0.05$. All analyses were performed at least in triplicate, and the experimental results were expressed as mean ± standard deviation (SD) at $p < 0.05$ (Loi and Tung, 2019).

3. Results and discussion

3.1 Some biochemical and physicochemical indexes of fresh pears before storage

Before putting pears into storage, analyze a number of biochemical and physicochemical indexes of fresh pears before putting them into storage to serve as a basis for determining the quality variation of this fruit during storage. The results are shown in Table 2.

Table 2. Some biochemical and physiological indexes of fresh pears before being put into storage.

TT	Some are just biochemical and biochemical.	Results
1	Soluble dry matter (°Bx)	11.52±0.13
2	Vitamin A (µg %)	41.36±0.17
3	Total sugar content (%)	9.83±0.12
4	Total Protein (%)	0.75±0.04
5	Respiratory Intensity (mL CO_2/kg/h)	89.37±0.18
6	Color variation	1.54±0.09
7	Fruit weight (g)	420±30

According to the results in Table 2, the fresh pear has a soluble dry matter content of 11.52°Bx, a vitamin C content of 41.36 mg %, a total sugar content of 9.83%, a total protein content of 0.75%, a respiratory intensity of 89.37 (mL CO_2/kg/h), a color variation of 1.54, and an average weight of 400-450 g. The pear is spherical and evenly rounded; the ripe skin has dark brown spots on a yellowish brown background; the height of the fruit is 65 -72 mm. The results of this study are also consistent with the results of the studies by the authors Francisco *et al.* (2017) and Shehzad *et al.* (2015).

3.2 The influence of the preservation membrane on the transformation of the biochemical indexes of pear

3.2.1 The effects of the preservation film on the variation in the content of soluble dry matter

Depending on the different storage conditions, the content of pear solubility changes quickly or slowly. The results of determining the variation in the dissolved dry matter content of pears during storage are shown in Table 3.

The dissolved dry matter content of the pear in the first week of storage was 11.63 (°Bx), the CT-2 formula was 11.23 (°Bx), the CT-3 formula was 11.08 (°Bx), the CT-4 formula was 11.31 (°Bx), and the CT-5 formula was 11.56 (°Bx). By the second week of storage, the content of the dried solute matter of the preserved pear in the experimental formulations using a modified preservation film was not much compared to the original. By the fourth week of storage, the content of pear solubility in formula CT-3 was 11.94 (°Bx) and formula CT-4 was 12.07 (°Bx). By week 4, all of the pears in the CT-5 formula have gone bad. This shows that the preserved pear in the CT-5 formula is covered with a thick film, with a large content of film-forming compositions that prevent water from escaping and depositing on the surface of the fruit shell, causing rapid deterioration. Thus, with a probiotic content of 1.0% in the CT-3 formulation, it was better to limit the variation in the content of soluble dry matter in pears than other experimental formulations.

Table 3. The effect of the preservative membrane on the variation of dissolved dry matter content.

Storage period (weeks)	Dissolved matter content variation (°Bx)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	11.63^a	11.23^d	11.08^e	11.31^c	11.56^b
2	-	11.89^a	11.23^c	11.52^b	11.87^a
3	-	12.76^a	11.45^d	11.86^c	11.96^b
4	-	-	11.94^b	12.07^a	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

3.2.2 The effects of the preservation membrane on the variation of vitamin C content

Vitamin C is an important micronutrient in vegetables in general and pears in particular, but vitamin C is often easy to lose during processing and storage. The results of determining the variation in vitamin C content of pears during storage are shown in Table 4.

Table 4. The effect of the preservation membrane on the variation of vitamin C content.

Storage period (weeks)	Variation of vitamin C content (mg%)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	36.32^d	40.85^b	41.23^a	40.74^b	40.25^c
2	-	36.25^d	40.16^a	39.35^b	38.76^c
3	-	-	38.59^a	36.42^b	34.29^c
4	-	-	35.74^a	29.86^b	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

The results in Table 4 show that during storage, the vitamin C content of pears gradually decreased in all experimental formulations. The pear in the CT-1 formula had a greater reduction in vitamin C content than the pear in the other experimental formulations. After a week of storing the first pear in the CT-1 formula, the remaining vitamin C content is 36.32 mg %. The vitamin

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C content of pears in the formulas CT-2, CT-3, CT-4, and CT-5 all remained higher than 40 mg % after a week of storage. In formulations using biofilms to preserve pears, the CT-3 formula had the lowest reduction in vitamin C content; by the fourth week of storage, the remaining vitamin C content was 35.74 mg %. Thus, the preservation film has the effect of effectively limiting the loss of vitamin C content in pears during storage. Because pears in CT-4 and CT-5 formulations are covered with a thick film, the composition's high content prevents water from escaping and depositing on the surface of the fruit peel, causing rapid deterioration and lowering vitamin C content more than pears in CT-3 formulation. The pear in the CT-2 formula loses more vitamin C content than the CT-3 formula because, with a probiotic content of 0.5%, it is not enough to limit the loss of vitamin C content. The results of this study are also consistent with the research results of Lysiak *et al.* (2021). That said, with a probiotic content of 1.0% in the CT-3 formulation, it effectively limits the variation in vitamin C content of pears during storage compared to other experimental formulations.

3.2.3 The effect of the preservation film on the variation of the total sugar content

When the pear is ripe, the sugar content increases due to starch metabolism, and tannin metabolism, which form sugar under the action of enzymes. However, during the preservation process, sugar content tends to decrease gradually due to being transformed into simpler substances. The results of determining the influence of the preservation film on the variation of the total sugar content of the pear are shown in Table 5.

Table 5. The effect of the preservative film on the variation of total sugar content.

Storage period (weeks)	Variation of total sugar content (%)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	7.67^{d}	8.26^{c}	9.34^{a}	9.07^{b}	9.03^{b}
2	-	6.74^{d}	8.27^{a}	7.93^{b}	7.82^{c}
3	-	-	7.42^{a}	6.87^{b}	6.28^{c}
4	-	-	7.25^{a}	6.13^{b}	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

After a week of preservation, the total sugar content of pears has not changed much in formulations using biofilms for preservation. However, during this time, the total sugar content of the pear in the CT-1 formula changed rapidly, and by the end of the first week of storage, it was completely rotten. By the fourth week of storage, only the preserved pears in the CT-3 and CT-4 formulas had not completely decayed. Comparing the variation in the total sugar content of the pears in these two formulas, the results showed that the preserved pears in the CT-3 formula had the lowest variation; specifically, the total sugar content changed from 9.34% to 7.25%, and the preserved pears in the CT-4 formula had a total sugar content change from 9.07% to 6.13%. Compared with the research results of Lysiak *et al.* (2021), this research result is completely similar. Thus, the preservative content of 1.0% in the CT-3 formulation effectively limited the total sugar content of the pear compared to other experimental formulations.

3.2.4 The effect of the preservation membrane on the change in total protein content

The total protein content of pears tends to decrease during storage due to degradation into simpler substances under the action of enzymes. The variation in the total protein content of the pear is shown in Table 6.

Table 6. The effect of the preservation membrane on the variation of the total protein content.

Storage period (weeks)	Total Protein Content Variation (%)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	0.64^{d}	0.67^{c}	0.72^{a}	0.71^{b}	0.69^{b}
2	-	0.54^{c}	0.68^{a}	0.63^{b}	0.54^{c}
3	-	-	0.63^{a}	0.59^{b}	0.53^{c}
4	-	-	0.57^{a}	0.52^{b}	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

The results of the study in Table 6 show that, after the first week of storage, the total protein content of pears in the CT-3 and CT-4 formulations had negligible variation compared to fresh pears. Meanwhile, pears preserved in the CT-1 formulas, CT-2 formulas, and CT-5 formulas all had protein content losses of 0.06 - 0.11% compared to fresh pears. The reason for this phenomenon is that the preserved pear in the CT-1 formula does not use a film-forming composition for preservation, while the CT-2 formula uses a film-forming composition of 0.5% that is unable to inhibit the activity of microorganisms and enzymes, leading to a large loss of total protein content. The pear preserved in the CT-5 formula is covered with a thick film, with a large film content that prevents water from escaping and depositing on the surface of the fruit peel, causing rapid rot and reducing the protein content more than the pear preserved in the CT-3 formula. Comparing the total protein content variation of the preserved pear in the CT-3 formula and the CT-4 formula, the results showed that the preserved pear in the CT-3 formula had the lowest protein content variation. The results of this study are also consistent with the research results of Lysiak *et al.*

(2021). That shows that with a probiotic content of 1.0%, it has the effect of limiting the transformation of the total protein content of pears more effectively than other composition contents.

3.3 The influence of the preservation film on the transformation of the physicochemical indexes of pears

3.3.1 The effects of the preservation membrane on the variation of respiratory intensity

Respiration is a complex physiological and biochemical process that transforms the organic substances in pears, which consume nutrients to form compounds that nourish and maintain the living activities of pears and partly radiate heat to the environment. The results of determining the influence of the preservation membrane on the variation in respiratory intensity of pears are shown in Table 7.

Table 7. The effects of the preservation membrane on the variation of respiratory intensity.

Storage period (weeks)	Variability of respiratory intensity (mL CO_2/kg.h)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	86.35^{a}	84.53^{c}	82,24^{e}	83.56^{d}	85.27^{b}
2	-	82.41^{a}	75.87^{d}	75.94^{c}	81.42^{b}
3	-	76.38^{a}	69,69^{d}	71.37^{c}	76.28^{b}
4	-	-	64.75^{b}	68.82^{a}	-

Values with different superscripts within the same row are statistically significantly different ($P<0.05$).

Table 7 shows that, in the first week of storage, the respiratory strength of pears preserved in the CT-1 formula is the strongest compared to pears in other experimental formulations. However, by the second week of preservation, the respiratory intensity of pears in preservation formulas gradually decreased, especially in those using probiotics for preservation. That shows that biofilms have effectively suppressed the respiratory intensity of pears in these formulations. By the fourth week of storage, the respiratory intensity of pears stored in the CT-3 formula decreased from 82.24 mL CO_2/kg/h to 64.75 mL CO_2/kg/h, and in the CT-4 formula, it decreased from 83.56 mL CO_2/kg/h to 68.82 mL CO_2/kg/h. By week 4, all of the pears in the CT-5 formula have completely rotten. That shows that the preserved pear in the CT-5 formula is covered with a thick film, with a large content of film-forming compositions that prevent water from escaping and depositing on the surface of the fruit shell, causing rapid deterioration. When the experimental formulations were compared, the results revealed that the CT-3 formulation's 1.0% film-forming probiotic content was more effective in suppressing respiratory intensity than the other experimental formulations.

3.3.2 The effects of the preservation film on color variation

During the preservation process, the color of the pear is always changed, affecting the quality of the fruit. The color variation of the pear during storage is shown in Table 8.

Table 8. The effect of the preservation film on the color variation.

Storage period (weeks)	Color variation				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	2.36^{a}	1.97^{c}	1.62^{e}	1.73^{d}	2.04^{b}
2	-	3.74^{b}	2.16^{d}	3.52^{c}	4.02^{a}
3	-	8.25^{a}	4.37^{d}	7.34^{c}	8.16^{b}
4	-	-	5.14^{b}	8.07^{ad}	-

Values with different superscripts within the same row are statistically significantly different ($P<0.05$).

The results in Table 8 show that, by the end of the first week of storage, the color change of the pear in the CT-1 formula is the largest, turning the fruit peel from its characteristic color to a dark brown. By the second week, the preserved pears in this formula have completely rotted. Meanwhile, the color of the pear preserved in the CT-2 formulation, CT-3 formulation, CT-4 formulation, and CT-5 formulation takes place more slowly, but the pear still has its characteristic color and peel. Thereby showing that the preservation film has the effect of effectively limiting the color variation of pears. By the second week of storage, the color variation of pears in these formulas tends to increase, especially in the CT-2 and CT-5 formulas. Comparing CT-3 and CT-4 formulations, the results showed that the preserved pear in the CT-3 formulation (with 1.0% film-forming probiotic content) had a slower color change than other formulations.

3.3.3 The effects of the preservation film on the loss of natural mass

The evaporation of water, a respiratory process that attracts the loss of nutrients and makes the skin dry and wrinkled, is primarily responsible for the loss of natural mass in pears. The goal of the storage process is to limit the loss of the natural mass of the pear in addition to limiting rot. The results of determining the effect of the preservation film on the loss of natural mass in pears are shown in Table 9.

Table 9 shows that, after a week of preservation, the loss of natural mass of pears in the CT-1 formula is the largest at 4.56%, followed by the CT-2 formula at 4.03%, and the CT-5 at 3.73%. Meanwhile, pears preserved in the CT-3 and CT-4 formulations had lower natural mass loss. This process occurs because the pear is

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stored in the CT-1 formula so that under natural conditions it is not covered with a preservation film, leading to water and other components evaporating and causing a large loss of natural mass. Preserved pears in the CT-5 formulation, due to being covered with too thick a preservation film, lead to strong anaerobic respiration that consumes large nutritional components to release CO_2 and energy, causing a higher loss of natural mass than pears in the CT-3 and CT-4 formulations. The preserved pear in the CT-2 formula, with its low content of preservative formulation, is not capable of inhibiting the action of microorganisms and enzymes, not enough to inhibit respiration and evaporation, as well as other nutritional components, leading to a large loss of natural mass. Comparing CT-3 and CT-4, pears preserved in CT -3 with a probiotic content of 1.0% had lower natural mass loss. Compared with the research results of Lysiak *et al.* (2021), this research result is completely similar. Thus, with a probiotic content of 1.0%, it has an effective effect of limiting the loss of natural mass, limiting water loss, and reducing the drying and wrinkling of pears during storage.

Table 9. The effect of the preservation film on the loss of natural mass.

Storage period (weeks)	Loss of natural mass (%)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	4.56[a]	4.03[b]	2.56[e]	2.84[d]	3.73[c]
2	-	6.46[a]	3.31[d]	4.27[c]	4.91[b]
3	-	8.74[a]	4.54[d]	5.48[c]	6.36[b]
4	-	-	5.23[b]	6.79[a]	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

3.3.4 The effect of the preservation film on the rate of deterioration

The purpose of the storage process is to limit spoilage and prolong the shelf life of the pears. The results of determining the influence of the preservation film on the rot rate of pears are shown in Table 10.

After a week of pear storage in all formulations, rotting occurs, with the highest rate of rotting occurring in the CT-1 formulation, followed by the CT-2 and CT-5 formulations. The pear preserved in the CT-1 formula is completely rotten by the second week of storage. There were only two CT-3 formulations by the fourth week of storage, and the pear formulation in the CT-4 formulation was not completely rotten, so the rate of rotten pears in the CT-3 formulation was lower than that in the CT-4 formulation. The reason why pears in CT-2, CT-4, and CT-5 formulas have a greater rate of deterioration than CT-3 is because CT-2 formulas with a low content of probiotics do not optimally inhibit the activity of microorganisms and enzymes, thus leading to pears rotting quickly. Formulas CT-4 and CT-5 with a high content of probiotics and a large thickness of preservation film cause water that escapes from the fruit shell to be prevented from being deposited on the surface of the shell, causing the fruit to rot quickly. The results of this study are also consistent with the research results of Lysiak *et al.* (2021). Thus, with a probiotic content of 1.0% in the CT-3 formulation, it effectively limited the rate of rotten pear growth compared with other experimental formulations.

Table 10. The effect of the preservation film on the rate of deterioration.

Storage period (weeks)	Corruption rate (%)				
	CT-1	CT-2	CT-3	CT-4	CT-5
1	23.64[a]	3.68[b]	1.24[e]	2.87[d]	3.56[c]
2	-	9.13[a]	2.18[d]	4.64[c]	8.74[b]
3	-	18.54[b]	3,36[d]	5.28[c]	19.67[a]
4	-	-	5.07[b]	6.15[a]	-

Values with different superscripts within the same row are statistically significantly different (P<0.05).

4. Conclusion

From the results described above, some conclusions are given as follows: Formulated chitosan combined with vinegar, sodium alginate, and carboxymethyl cellulose at the rate of 160 g chitosan/160 mL vinegar/7,000 mL water, 4 g sodium alginate, and 0.5 g carboxymethyl cellulose to preserve 180 kg of pears. Since then, five pear preservative formulations have been developed: CT-1 (control formulation, not using preservative film formulation), CT-2 (using 0.5% preservative film formulation), CT-3 (using 1.0% preservative film formulation), CT-4 (using 1.5% preservative film formulation), and CT-5 (using 2.0% preservative film formulation). In 5 preservation test formulations, it was determined that the content of probiotics at 1.0% is suitable for preserving pears. When stored in the content of this probiotic, it has limited the transformation of biochemical and physicochemical indexes. Especially by the fourth week of storage, pears still retain their characteristic values; the natural loss rate is only 5.23%, and the rot rate is 5.07%, both lower than when stored in other biological products. Compared with fresh pears before being put into storage, the change in biochemical and physicochemical indexes of preserved pears with a biological content of 1.0% is not large. Therefore, choosing the CT-3 formula, with a probiotic content of 1.0%, to build a pear preservation process is appropriate.

Conflict of interest

The author declares the research results in this article to be completely honest. The data has never been used or rotated from other research projects in any form.

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