Effects of edible coatings of chitosan - fish skin gelatine containing black tea extract on quality of minimally processed papaya during refrigerated storage

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Effects of edible coatings of chitosan - fish skin gelatine containing black tea extract on quality of minimally processed papaya during refrigerated storage

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ABSTRACT

Papaya fruit is a climacteric fruit that undergoes physical and chemical changes very quickly during storage. The edible coating is one method that can extend the shelf life of minimally processed fruit such as papaya. To improve the effect of edible the mechanical properties and functional value of edible coatings, black tea was added. The research aims to evaluate the application of edible coating from fish gelatin-chitosan enriched with black to the physicochemical characterization of the minimally processed papaya.

The research was conducted through the extraction of tuna skin gelatin using the acid-base method. Furthermore, the preparation of edible coating solutions based on chitosan, gelatin, glycerol, and black tea with different concentrations of 0%, 5%, 10%, and 15%. The edible coating solution that has been obtained is then applied to minimally processed papaya fruit and stored for 1, 4, 7, and 10 days to determine the effect of edible coating on the physical, chemical and microbiological characteristics of papaya fruit.

The results showed that the addition of black tea extract concentrations of 0%, 5%, 10%, and 15% had no significant effect on the color change of the resulting solution (p > 0.05), while the viscosity and pH occurred during the addition of the concentration to the coating solution which could eat (p < 0.05). Edible coating can suppress the decrease in weight loss and papaya texture. The chemical properties of minimally processed papaya such as pH, total dissolved solids, total titrated acid, antioxidant activity, and total carotenoids increased due to the ripening process and the effect of dissolved substances contained in the edible layer during storage. Total plate count (TPC) showed that edible coating treatment with the addition of black tea extract was able to suppress microbial growth during storage.

1. Introduction

Minimally processing fruits or also known as fresh-cut fruits is the processing of fruit that involves washing, peeling, and slicing before being packaged and using low temperatures for storage without losing their freshness and nutritional value (Poverenov et al., 2014). One of the minimally processed fruits that are commonly found in the market is papaya. Papaya (*Carica papaya* L.) is one of the fruits that can be processed minimally. Papaya flesh has a sweet taste, is soft, and has a lot of

water and carbohydrate content. Papaya is an excellent source of carotene, vitamins, protein, and polysaccharides (Polido et al., 2021). On the other hand, papaya is a climacteric fruit that undergoes very rapid physical and chemical changes due to differential gene expression, enzyme, and ethylene activation (Jafarzadeh et al., 2021). Climacteric fruit increases respiration rate and ethylene production which results in decreased fruit quality during storage. Changes in papaya fruit during storage include soft texture, reduced fruit weight, decreased nutritional value, and increased microbial growth (Salehi, 2020). To prevent

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damage and spoilage of the fruit, a handling method is needed that can ensure that consumers can enjoy fresh papaya fruit of good quality and quality.

One method that can extend the shelf life of minimally processed fruit is packaging with edible coatings (Brasil et al., 2012). The edible coating is defined as a thin layer that covers the surface of food and can be eaten as part of products made from natural ingredients such as polysaccharides, lipids, and proteins that are environmentally friendly (Yıldırım-Yalçın et al., 2021). Sources of edible coating biopolymers include polysaccharides (starch, alginate, carrageenan, chitosan), protein (gelatin, casein, collagen, gluten, soy), and lipids (fatty acids, waxes, resins) (Rodrigues et al., 2020). The application of edible coatings from aloe vera on cut papaya fruit showed that the coating was able to extend the shelf life of fresh-cut papaya (Farina et al., 2020). The coating can reduce the loss of vitamin C and carotenoid content. Based on these studies, chitosan and gelatin can play a role in inhibiting damage to fresh-cut papaya.

Gelatin exhibits good barrier characteristics against oxygen transfer and aroma. However, gelatin has poor barrier properties against water vapor transfer due to its hydrophilic nature so the addition of other substances can be used to improve water vapor transfer properties (Ningrum et al., 2021). In addition, chitosan and its derivatives have various advantages as matrix materials, such as having antibacterial and antioxidant activity and increasing mechanical strength (Sahraee et al., 2019). Therefore, the addition of antimicrobial and antioxidant agents can also impair the mechanical properties and enrich the functional value of edible coating on papaya fruit. Previous research also showed the application of composite fish gelatin with chitosan can improve the quality of minimally processed fruit such as watermelon (Salsabiela et al., 2022).

Previous research conducted showed that the application of the edible coating on papaya fruit using Aloe vera gel showed that the Aloe vera layer acts as a physical barrier for water loss and respiration thereby reducing weight loss, controlling pathogenic microbes in papaya fruit, and able to delay fruit ripening (Farina et al., 2020). Other additives that can be used to add functional value are green tea extract and pu-erh (Camellia sinesis L.) which have been widely used in the food industry. Tea extracts are most well-known for their high antioxidant properties, but can also be used as antimicrobial agents (Giménez et al., 2013), and tea polyphenols also exhibit inhibitory effects on starch retrogradation (Sahraee et al., 2019). In addition, black tea contains antioxidants that play a role in cross-linking agents for coating manufacture. Tea extract can improve film components which is confirmed by the improvement of mechanical properties, and antioxidant and antimicrobial activity (Hosseini & Gómez-Guillén, 2018). Based on our previous study, black tea contains tannins such as thearubigins, which have antioxidant and antimicrobial activities in edible films (Salsabiela et al., 2022). On the other hand, several polyphenolic compounds, such as catechins, theaflavins, and methyl jasmonate, also have antimicrobial and antioxidant activity (Salsabiela et al., 2022). The research aimed to investigate the effect of the incorporation of black tea into the active film of chitosan-gelatin derived from tuna skin on its mechanical, antioxidant, and antibacterial properties. Likewise, the potential use of the developed edible coating in improving the shelf life of processes papaya fruit was also evaluated to provide a future option for developing sustainable and environmentally friendly active film in the fruits industries especially minimally processing papaya fruits.

2. Materials and methods

2.1. Materials

The material used was papaya, black tea (TongJi, Indonesia) tuna skin, 0.2% sodium hydroxide (NaOH) (Merck, Germany), and 0.2% sulfuric acid (H₂SO₄) (Merck, Germany), citric acid (Merck, Germany). C₆H₈O₇) (Merck, Germany), DPPH (2,2-diphenyl-1-picrylhydrazyl

hydrate) (Sigma Aldrich, Singapore), ethanol PA (Merck, Germany), distilled water, Whatman No. filter paper. 1, silica gel, aluminum foil, filter cloth, Place Count Agar (PCA) (Merck, Germany), and NaCl (Merck, Germany).

2.2. Extraction of gelatin from tuna fish skin

The extraction method with pre-treatment is based on the method of Tkaczewska et al. (2018). The skin of the tuna is moistened and then immersed in sodium hydroxide for 2 h with a sample ratio of 1: 6 (w / v) of alkaline. Furthermore, the sample is washed with distilled water until it reaches a pH of 6-7. Then the sample was immersed in sulfuric acid for 2 h with a ratio of sample and acid 1: 6 (w / v) then the sample was washed with distilled water until it reached a pH of 6-7. Furthermore, the sample was immersed in citric acid for 2 h with a ratio of sample and citric acid 1: 6 (w / v) and then washed with distilled water until it reached a pH of 6-7. Then the sample was extracted using distilled water with a ratio of 1: 3 (w / v) at a temperature of 45 \pm 1.5 °C for 60 min. After the extraction process is complete, the sample is filtered using a filter cloth. Then the sample is dried and ground to obtain gelatin powder.

2.3. Preparation of edible coatings

The preparation of a tea extract solution, namely dry tea powder, was added with distilled water at a ratio of m/v 1: 100 at 90°C for 2 min. The extract was then filtered with Whatman No.1 paper. The next is the process of making a coating solution. The tuna skin gelatin powder that has been obtained is then dissolved by adding distilled water and stirring at 40 °C for 30 min. After obtaining the gelatin solution, a 1.5% concentration of chitosan was added which was dissolved in acetic acid and distilled water. In addition, glycerol is added which acts as a plasticizer. The addition of a glycerol plasticizer can increase flexibility. After the ingredients were dissolved, black tea extract was added according to the predetermined concentrations, 0%, 5%, 10%, and 15%. After adding the black tea extract, it was stirred again for 15 min and an edible coating solution was obtained.

2.4. Characterization of coating solutions

2.4.1. Colour

Colour intensity of edible coating solution was measured using a Chromameter Minolta CR-400. The Minolta CR-400 Chromameter uses L, a, and b systems. The display will display the values of L, a, and b in 4 numbers each. The test is carried out by attaching the sensor to the coating solution and firing a beam at the part to be measured (Ningrum et al., 2020). We used The Chroma Meter that has aperture 0.8 cm and the white calibration plate/standard observer was used for a calibration prior to sample analysis (Y = 86.51 x = 0.3168 and y = 0.3245); illuminant D65, °observer 10° .

2.4.2. Viscosity of coating solutions

Brookfield viscometer is a viscometer that uses a spindle dipped in the substance being tested and measures the test substance and measures the resistance to motion of the rotating part. The viscosity of the coating solution was measured at a temperature of 25 °C and a speed of 60 rpm.

2.4.3. pH of coating solutions

Testing the pH for each sample using a digital pH meter. The pH of coating solution samples was tested at a temperature of 25 $^{\circ}$ C (Mendy et al., 2019).

2.5. Physicochemical properties

2.5.1. Colour

The Colour intensity of the fresh cut papaya sample was measured



using a Chromameter Minolta CR-400. The Minolta CR-400 Chromameter uses L, a, and b systems. The display will display the values of L, a, and b in 4 numbers each. The test is carried out by attaching the sensor to the papaya sample and firing a beam at the part to be measured (Ningrum et al., 2020).

2.5.2. Texture

Hardness measurements were carried out using a universal texture machine (Zwick | Roell Z0.5, Germany). The hardness test was measured based on the level of resistance of the fruit to the rheometer needle. The measurement is set to mode 20, the maximum load is 2 kg, the pressing depth is 10 mm, the load drop speed is 30 mm/min and the needle diameter is 5 mm. Each measurement was repeated twice per test sample. The test was carried out in the middle of the cut fruit (Mendy et al., 2019).

2.5.3. Weight loss

Weight loss is one of the factors that indicate the quality of papaya fruit. The weight of the fruit was weighed using an analytical balance. Weight loss was obtained from the difference between the initial weight of papaya and the final weight during storage (Mendy et al., 2019).

The calculation of weight loss were presented below:

Weight loss (%) =
$$\frac{\text{initial weight (g) - final weight (g)} \times 100\%}{\text{Initial weight (g)}}$$

2.5.4. Total soluble solids

Measurement of total soluble solids (TSS) was determined by using a digital refractometer brand Atago PR-210 type with a resolution of 0.1% Brix and an accuracy of \pm 0.2% Brix. A refractometer is a simple optical instrument that measures the amount of light left in a liquid and checks for brix. Papaya flesh is taken, cut, and then squeezed to get the filtrate which is then placed on the refractometer lens to read the results. Each time measurement, the lens is cleaned using distilled water and calibrated before starting again. pH was determined using a pH meter (Model GLP 21). The juice from soluble solid concentration was used to determine pH. The pH meter was calibrated with buffers at pH 7.0 and 4.0 before being used (Mendy et al., 2019).

2.5.5. pH

Testing the pH for each sample using a digital pH meter. A sample of 10 g of papaya was homogenized with 100 mL of distilled water and the results were recorded daily and expressed as pH units (Mendy et al., 2019).

2.5.6. Titratable acidity

Titratable acidity was evaluated following AOAC method 942.15A, in which 5 mL of diluted papaya juice in 95 mL distilled water was titrated, with 0.1 M NaOH, and expressed as a citric acid percentage (AOAC, 1998). The endpoint of the titration is indicated by the formation of a stable pink color.

2.5.7. Total carotenoids

Papaya pulp (5 g) was grounded in the mortar with 10 mL of extraction solution (hexane: acetone: ethanol; toluene at 10: 7: 6: 7 v/v/v/v/v). The extracts were transferred into test tubes after which 1 mL of methanolic KOH was added and heated in a water bath at 56°C for 20 min for saponification. It was immediately cooled at room temperature before 10 mL of 10% sodium sulfate was added and stirred for 1 min. After the phase separation, the upper phase was used for carotenoid analysis using a spectrophotometer UV-VIS at 450 nm (Mendy et al., 2019).

2.5.8. Antioxidant activity

DPPH (2, 2-diphenyl-1-pycrylhydrazyl) scavenging activity of papaya fruits was determined by the modified method of with a slight

Table 1
Colours of black tea extract edible coating solution.

Black Tea Extract Concentration	Colours Lightness	a*	b*
0%	25.69 ± 2.22^a	0.22±0.25 a	1.16±0.16 a
5%	23.8±1.49 a	0.04 ± 0.42^{a}	1.02±0.06 a
10%	23.79 ± 1.26 a	0.22 ± 0.46^{a}	1.08 ± 0.21^{a}
15%	23.32 ± 0.61 a	$0.15{\pm}0.41~^a$	$1.02\pm0.22^{\ a}$

Notes: The different letter notations (a, b, c,) indicate a significant difference in the concentration of black tea extract treatment between samples (P < 0.05)

modification (Mendy et al., 2019). About 1 mL of papaya samples was mixed thoroughly with 3 mL freshly prepared DPPH solution and kept in the dark for 30 min at room temperature before the absorbance was read at 517 nm using UV-VIS spectrophotometer (Fisher Thermo Scientific Multiskan Go Model 151, United Kingdom). The percentage of DPPH scavenging activity was determined by the following equation (Mendy et al., 2019).

$$\textit{DPPH radical scavenging activity} \ (\%) = \frac{\textit{A blanko} - \textit{A sample}}{\textit{A blanko}} x \ 100\%$$

2.6. Total plate count (TPC)

Papaya fruit samples were tested using the pour method. In the plate count method using the pour plate method, 1 mL of the sample (all dilution series) is taken and poured into a sterile petri dish. Then pour 10 mL of PCA media into the petri dish. After that, the media was flattened by shaking the petri dish and incubated at 30 °C for 24 to 48 h (Sipahi et al., 2013)

2.7. Experimental design and statistical analysis

The study involved a two-factorial arrangement of treatments with four different concentrations (0%, 5%, 10%, and 15%, v/v) x storage durations (1, 4, 7, and 10 days), with three replications. Data obtained were subjected to analysis of variance (ANOVA) and Duncan's multiple range test (DMRT) was used to compare the means at 95% confidence level.

3. Results and discussion

3.1. Characterization of coating solutions

3.1.1. Colour

The results of statistical tests showed that the addition of extracts had no significant effect to the, a and b values of the coating solution (p>0.05) (Table 1). Changes in the degree of L*, a*, and b* values were related to the enzymatic oxidation treatment because this enzymatic oxidation played a role in changing the content of tannin compounds into theaflavins and thearubigins. Theaflavins play a role in determining the brightness of the tea steeping colour (reddish yellow). Thearubigin is a compound that is difficult to dissolve in water and plays a role in determining the stability of the color of steeping tea (dark brownish red) (Rohdiana, 2006). In addition, in the enzymatic oxidation process,

Table 2
Viscosity and pH of black tea extract edible coating solution.

Black Tea Extract Concentration	Parameter Viscosity	pН
0%	215.82±32.5°	4.25±0.06 ^a
5%	187.5 ± 22.98^{bc}	4.38 ± 0.04^{b}
10%	154 ± 16.37^{b}	4.57 ± 0.10^{c}
15%	114.17 ± 25.87^{a}	4.65 ± 0.14^{c}

Notes: The different letter notations (a, b, c,) indicate a significant difference in the concentration of black tea extract treatment between samples (P < 0.05)

Table 3

Colors of papaya with a minimal edible coating of black tea extract during storage.

110				
Black Tea Extract	Storage (Day	y)		
Concentration	1	4	7	10
Lightness (L)				
Control	47.30	44.37	44.24	42.38
	$\pm 2.59^{aB}$	$\pm 2.87^{aB}$	$\pm 1.83^{aB}$	$\pm 4.27^{aB}$
0%	48.15	45.62	46.30	46.31
	$\pm 2.79^{aA}$	$\pm 0.45^{aA}$	$\pm 1.42^{aA}$	$\pm 1.52^{aA}$
5%	45.33	46.45	43.63	45.01
	$\pm 2.06^{aA}$	$\pm 1.34^{aA}$	$\pm 3.46^{aA}$	$\pm 1.01^{aA}$
10%	45.43	43.07	44.32	45.96
	$\pm 0.61^{aA}$	$\pm 2.31^{aA}$	$\pm 1.92^{aA}$	$\pm 2.13^{aA}$
15%	44.43	47.83	45.40	44.02
	$\pm 0.82^{aA}$	$\pm 0.84^{aA}$	$\pm 1.57^{aA}$	$\pm 3.56^{aA}$
a				
Control	20.03	12.78	9.58	7.99
	$\pm 1.97^{aB}$	$\pm 4.02^{aB}$	$\pm 5.36^{aB}$	$\pm 3.10^{aB}$
0%	17.99	12.88	12.93	11.94
	$\pm 1.36^{aA}$	$\pm 4.13^{aA}$	$\pm 2.39^{aA}$	$\pm 2.62^{aA}$
5%	19.33	14.71	12.13	12.15
	$\pm 0.91^{aA}$	$\pm 1.15^{aA}$	$\pm 0.41^{aA}$	$\pm 0.45^{aA}$
10%	20.31	11.22	9.31	14.20
	$\pm 0.64^{aA}$	$\pm 1.24^{aA}$	$\pm 1.37^{aA}$	$\pm 1.22^{aA}$
15%	20.26	11.67	11.08	11.40
	$\pm 0.50^{aA}$	$\pm 3.51^{aA}$	$\pm 1.35^{aA}$	$\pm 0.82^{aA}$
b				
Control	29.89	18.55	17.20	17.77
	$\pm 2.14^{aB}$	$\pm 4.38^{aB}$	$\pm 5.20^{aB}$	$\pm 4.57^{aB}$
0%	30.23	21.61	20.94	19.92
	$\pm 1.32^{abA}$	$\pm 0.90^{abA}$	$\pm 2.99^{abA}$	$\pm 1.75^{abA}$
5%	32.58	23.06	20.37	22.96
	$\pm 1.81^{bA}$	$\pm 1.98^{bA}$	$\pm 1.80^{\rm bA}$	$\pm 2.21^{hA}$
10%	25.37	19.22	22.81	21.64
	$\pm 2.47^{abA}$	$\pm 0.51^{abA}$	$\pm 1.52^{abA}$	$\pm 1.52^{abA}$
15%	28.21	23.44	21.64	20.46
	$\pm 1.96^{abA}$	$\pm 2.37^{abA}$	$\pm 2.65^{abA}$	$\pm 4.05^{abA}$

tannins are oxidized to theaflavins and condensed to thearubigin. These compounds are together responsible in contributing contribute the darker color of the tea (Ngure et al., 2009).

3.1.2. Viscosity of edible coating solution

Table 2 reports, that the highest viscosity is the concentration of 0% black tea extract is 215.81cp. The lowest viscosity is the concentration of 15% black tea extract of 114.16 cp. Meanwhile, the results of the variance test showed that the addition of black tea extract had a significant effect on the viscosity of the solution (p < 0.05). The viscosity indicates the viscosity of the solution. The viscosity of the solution is influenced by the type of raw material used. The decrease in viscosity indicates the weakening of the intermolecular forces in the edible coating solution. The intermolecular forces are related to the number of hydrogen bonds in the film solution. The addition of tea polyphenols can weaken the hydrogen bonding force due to the interaction between the benzene ring of tea polyphenols and chitosan (Liu et al., 2019). Particles with a higher charge have a greater viscosity due to the greater attraction of opposing ions, making droplet movement more difficult and the effective diameter is larger so that they cannot stick together due to electrostatic repulsion. The combination of gelatin-chitosan will be able to form polyelectrolytes which can improve the mechanical properties. In the gelatin-chitosan layer, an ionic bond is formed between the negative charge of the carboxylate group on the gelatin and the positive charge of the amine group on the chitosan to form an electrostatic complex or a polyelectrolyte complex.

3.1.3. pH of edible coating solution

The lowest pH was the concentration of 0% black tea extract or without the addition of black tea extract 4.24. While the highest pH is a concentration of 15% black tea extract of 4.64. Meanwhile, the results of the variance test showed that the addition of black tea extract had a significant effect on the viscosity of the solution (p < 0.05) (Table 2). The presence of alkali metals or bicarbonate salts is thought to be the cause of the high pH. The mechanical strength and stiffness of gelatin gels combined with polyphenols increased significantly. Gelatin-polyphenol crosslinking has significantly increased gel strength with a more compact surface. The interaction between phenolic compounds and proteins is not only influenced by the structure of the phenolic compounds but also by the surface properties of the protein. Different proteins have different amino acid compositions, isoelectric points, and hydrophobic interactions, which have an impact on the cross-linking ability of proteins with phenolic compounds (Hosseini Gómez-Guillén, 2018). In addition, what affects the pH of the coating solution is the quality of black tea which is determined by the number of theaflavins under storage conditions. The number of theaflavins will decrease if stored at low temperatures, low humidity levels, and low oxygen availability as well. The remaining activity of the peroxidase enzyme will also accelerate the decrease in the amount of theaflavin during storage. The degree and speed of oxidation of theaflavins in tea depend on the pH of the water used for brewing.

3.2. Physicochemical properties

3.2.18 Colour

Color is considered one of the most important and crucial attributes associated with fresh fruit such as papaya (Polido et al., 2021). This property exerts great influence on the acceptability of the product by the consumer and for this reason the color of edible coatings applied on papaya fruit surface did not provoke undesirable visible change in the food appearance Table 3 shows, that the value of L* (Lightness) decreased during storage for all treatments. On the control sample on day 1 of 47.30 it was lower on day 10 of 42.38. Edible coating samples with the addition of black tea can maintain the brightness value, such as at a concentration of 5%. With the addition of 5% black tea extract, the L value on the 1st day was 45.33 and increased on the 4th day by 46.44 which could be influenced by the presence of a layer, and then decreased until the 10th day by 45.01. The statistical test results showed that the coating treatment had no significant effect on the level of change in the L* value (p>0.05), while during the storage period, there was a significant effect between the 1st and 4th-day observations (p < 0.05). Previous research with the addition of bioactive extract in the edible coating of fresh-cut papaya during storage also showed a similar result. The darkness of minimally processed fruit during the storage period can be related to the action of polyphenol oxidase that in contact with the oxygen of the environment potentiates browning oxidative reactions. The use of edible coating must have minimized the contact between the surface of the papaya and oxygen, decreasing the oxidative browning of samples during storage. This effect can be attributed to pectin's capability to form strong chemical interactions with calcium ions resulting resistance network structure that difficult access oxygen to the food surface (Polido et al., 2021).

The results of the statistical test of the a^* value showed that the coating treatment had no significant effect on the rate of change in the a^* value (p>0.05), while during the storage period, there was a significant effect between the 1st and 4th-day observations (p < 0.05). The a^* value decreased significantly on the 10th day of observation, the degradation of carotenoids that occurred was accelerated by respiration which continued during storage. When compared with control fruit with coating treatment, the value of the change in color degree was low. This can be caused by the decomposition of the color component which is accelerated by the fruit's metabolic process during storage. Coatings containing polysaccharides can suppress metabolic activities that occur.

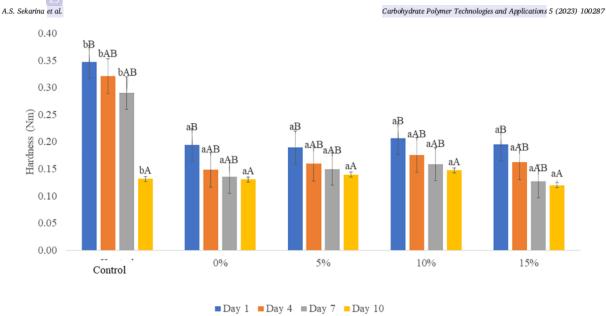


Fig. 1. Texture (Hardness) of papaya with minimal edible coating of black tea extract during storage Note: Different letter notations (A,B,C) indicate a significant difference in the treatment of storage days between samples (P < 0.05); while the different notation of letters (a,b,c) indicates a significant difference in the treatment of black tea extract concentrations between samples (P < 0.05).

This is also aligned with previous research on the application of polysaccharides coating on minimally processed papaya coated with alginate and modified atmosphere observed a decrease in the values of a value, and that the control sample presented major decreased for this parameter. These results agree with the present study, which also observed decreases in the a value over the days of storage for the control sample (Tabassum & Khan, 2020).

The degree of b* value indicates the yellow and red color of agricultural products caused by the presence of carotenoid pigments. The synthesis of these carotenoid compounds may have similarities to the formation of carotene and phytol, where the compounds released in the chlorophyll degradation process will be used for carotenoid synthesis. The yellow color in the sample with the addition of 0%, 5%, 10%, and 15% extracts has relatively the same level of yellow color. Changes in the b* value of papaya during storage based on statistical tests showed that the control treatment and edible coating with black tea extract had no significant effect on changes in the b value (p>0.05). In all samples, during the storage period, there was a significant effect on day 1 and day 4 (p < 0.05). The lowest decrease in b* value was the control treatment on the 1st day of 29.89 and the 10th day of 17.70. In addition, the lowest edible coating treatment was with black tea extract 0% on the 1st day of 30.22 and the 10th day of 19.91 (Table 3).

The amount of carotenoids formed in the degradation process of chlorophyll is greater than the amount of chlorophyll that is degraded. The degradation of chlorophyll causes the carotenoid pigments that were already present in the tissue to dominate the formation of a new color, namely yellow (Mukhaimin et al., 2019). The content of carotenoids, free geraniol, and free mevalonic acid, which are precursors for the formation of carotene, increases over time during ripening. The difference in the b* value indicates that the edible coating treatment was able to inhibit the decrease in the b* value in papaya fruit compared to the control. During storage, the color component of the fruit continues to change. The appereance of minimallyy processed papaya was shown in Fig. 1.

3.2.2. Texture

The decrease in hardness occurred very quickly during the storage

period in the control sample compared to the treatment with coatings. The hardness of control papaya fruit on day 1 was 0.35 Nm and on day 10 was 0.13 Nm. The lowest decrease in fruit hardness was found in papaya fruit samples with the addition of 5% black tea extract in the edible coating solution. The decrease on day 1 was 0.19 Nm and on day 10 was 0.14 Nm. The results of statistical tests showed a significant difference between the control sample and the edible coating treatment (p < 0.05). This comparison was due to the inhibition of the transpiration process due to the presence of a coating layer on minimally processed papaya fruit causing reduced water loss in the fruit so that the fruit hardness was higher than the control. In the papaya fruit sample, the difference in concentration of black tea extract between samples with edible coating treatment was not significantly different (p>0.05). The decrease in hardness of papaya fruit during storage had a significant difference on day 1 and day 10 (p < 0.05) (Fig. 1)

Hardness can be maintained at low-temperature storage because it causes the respiration process to decrease so that transpiration is low. The hardness level of fresh-cut papava fruit during storage tends to decrease. The decrease in the value of fruit hardness was caused by the process of respiration, transpiration, and microbial activity. Respiration is a process of reshuffling complex materials (starch, sugar, and organic acids) into simpler molecules (CO2, water, energy, and other new molecules) with the help of O2 that occurs in cells). Samples of papaya fruit were minimally affected by storage time and coating application (Salehi, 2020). The reshuffling of these complex materials can affect the texture of the fruit because the complex materials for building cell walls in the flesh of the fruit are broken down, causing the fruit texture to become soft and susceptible to mechanical damage. Fresh-cut papaya products will lose water content during storage so that cell turgor will decrease and result in decreased fruit hardness. Hardness loss (softening) is another characteristic of papaya maturation linked to enzymatic degradation of cell wall components and decomposition of intracellular materials (dos Passos Braga et al., 2020).

Fresh-cut agricultural produce is highly perishable as its exposed tissue becomes vulnerable to deterioration due to the lack of a protective

Table 4
Weight loss of papaya with minimal edible coating of black tea extract during storage.

Black Tea Extract Concentration	Storage (Day)	4	7	10
Weight loss (%)				
Control	0.321 ±0.131 abA	0.347 $\pm 0.147^{abB}$	0.29 ±0,043 ^{abC}	0.131 $\pm 0.044^{abD}$
0%	0.194 ±0.032 ^{aA}	0.131 ± 0.025^{aB}	0.136 ±0,042 ^{aC}	0.149 ±0,053 ^{aD}
5%	0.189 ±0.026 ^{hA}	0.14 ± 0.013^{bB}	0.15 ±0,038 ^{bC}	0.16 ±0,018 ^{bD}
10%	0.207 ±0.016 ^{abA}	0.175 ±0,04 ^{abB}	0.147 ±0,017 ^{abC}	0.159 ±0,005 ^{abD}
15%	0.196 ± 0.038^{aA}	0.12 ±0,011 ^{aB}	0.162 ±0,042 ^{aC}	0.127 ±0,023 ^{aD}

cover. Weight loss is considered an indicator of the freshness of fruits and vegetables (Tabassum & Khan, 2020). The weight loss of fresh-cut papaya was observed in all the samples.

In Table 4, the increase in weight loss began to occur from the 4th day to the 10th day of storage. The comparison of treatments showed that the control sample had a higher increase than the coating treatment. The weight loss of the control sample on day 4 was 1.88% and on day $10\,$ was 6.02%. Fruits that experienced the lowest increase in weight loss were those that were coated with the addition of 15% black tea extract concentration on the 4th day of 1.91% and 4.61% on the 10th day. The results of statistical tests on all samples showed a significant effect on coating samples with the addition of 5% black tea extract (p < 0.05). Changes in weight loss during all treatments during the storage period based on statistical tests had a significant or significant effect on each day of observation (p < 0.05). This result of weight loss of fruit during storage occurs because of evapotranspiration and respiration of the fruit and which is intensified with minimal processing treatment. Proteins such as gelatin and polysaccharides such as chitosan-based coatings tend to act as a barrier to water vapor in minimally processed fruits, reducing the mass loss during storage (Polido et al., 2021).

The higher the weight loss value, the higher the weight loss so that the weight of the fruit sample will decrease. Weight loss generally occurs due to loss of moisture or leakage of juice and during storage mainly due to respiration and transpiration of the produce, however, respiration is believed to contribute negligibly to weight as its products mostly include gases and aroma compounds (Tabassum & Khan, 2020). The decrease in fruit weight was caused by the fruit doing respiration by converting sugar into CO₂ and H₂O accompanied by the evaporation process of water vapor. This causes the percentage of weight loss to increase. The increase in weight loss occurred during the storage period. Storage at low temperatures causes reduced metabolic activity and chemical changes take place more slowly so that weight loss can be suppressed. A slow respiration rate will be able to slow down the water loss process so that the decrease in weight loss will also be lower (Salehi, 2020).

This result is in line with the parameters of the respiration rate and the decrease in the water content of the fruit. The respiration rate is the cause of the fruit experiencing a decrease in water content which will greatly affect the weight loss of the fruit. The substrate used for respiration every hour is at least 0.01% which will make agricultural products lose about 2-3% in weight at the recommended storage conditions (Salehi, 2020).

3.2.4. Total soluble solids

The highest increase in total soluble solids (TSS) was edible coating papaya fruit with the addition of 15% black tea extract of $8.83\,^{\rm o}$ Brix, while the lowest was the control sample of $8.33\,^{\rm o}$ Brix on the 10th day.

Table 5Total soluble solids, pH and titratable acidity of papaya with minimal edible coating of black tea extract during storage.

Black Tea Extract Concentration	Storage (Day)	4	7	10		
Concentration						
Total Soluble Solids (Brix)						
Control	7.75	7.833	8.25	8.333		
	$\pm 0.25^{aA}$	$\pm 0.144^{aA}$	$\pm 0.433^{aB}$	$\pm 0.946^{aB}$		
0%	7.75	7.917	8.083	8.5 ± 0.5^{aB}		
	± 0.433 aA	$\pm 0.382^{aA}$	$\pm 0.629^{aB}$			
5%	7.917	8.167	8.417 ± 0.52	8.667		
	± 0.144 aA	$\pm 0.289^{aA}$	aB	$\pm 0.52^{\ aB}$		
10%	7.5 ± 0.25 aA	8±0.5 aA	8.417	8.5 ± 0.5^{aB}		
			$\pm 0.382^{aB}$			
15%	7.75 ± 0.25	7.917	8.583	8.833		
	aA	$\pm 0.144^{aA}$	$\pm 0.144^{aB}$	$\pm 0.289^{aB}$		
pH						
Control	5.978	5.817	5.662	5.54		
	$\pm 0.687^{aC}$	$\pm 0.405^{aB}$	$\pm 0.288^{aAB}$	$\pm 0.23^{aA}$		
0%	5.853	5.65	5.627	5.577		
	±0.655 aC	± 0.565 aB	$\pm 0.443^{aAB}$	$\pm 0.388^{aA}$		
5%	5.81	5.693	5.618	5.517		
	±0.626 aC	± 0.56 aB	$\pm 0.438^{aAB}$	$\pm 0.333^{aA}$		
10%	5.873	5.697	5.505	5.407		
	±0.65 aC	$\pm 0.517^{aB}$	$\pm 0.251^{aAB}$	$\pm 0.174^{aA}$		
15%	5.86	5.708	5.512	5.313		
	±0.689 aC	$\pm 0.454^{aB}$	$\pm 0.292^{aAB}$	$\pm 0.114^{aA}$		
Titratable acidity (%)						
Control	0.255	0.285	0.321 20	0.345		
	$\pm 0.037^{aA}$	$\pm 0.041^{aA}$	±0.086 aA	$\pm 0.053^{aB}$		
0%	0.24	0.234	0.246	0.342		
	±0.037 aA	±0.036 ^{aA}	±0.061 aA	±0.018 ^{aB}		
5%	0.273	0.24±0.05	0.249	0.33±0.06		
	±0.092 aA	aA	±0.081 aA	aB		
10%	0.228	0.24	0.342	0.366		
	±0.027 aA	±0.073 aA	±0.047 aA	±0.063 ^{aB}		
15%	0.249	0.276	0.291	0.348		
	±0.045 aA	±0.044 ^{aA}	±0.087 aA	±0.101 aB		

Notes: The different letter notations (a, b, c,) indicate a significant difference in the concentration of black tea extract treatment between samples (P < 0.05); while different letter notations (A, B, \mathbb{C}) indicate a significant difference in storage day treatment between samples (P < 0.05)

The results of statistical tests showed that all treatments had no significant difference (p>0.05). Changes in TSS can be seen during the storage period on the 4th and 7th days there are significant differences (p < 0.05) (Table 5). TSS shows dissolved solids, pectin, and other organic acids (Salari et al. 2012). Changes in TSS during storage are one result of the continued fruit metabolism process (Yıldırım-Yalçın et al., 2021).

TSS of all the samples showed a gradually increasing trend during the entire storage period and a drastic change in the sugar content was not observed as also reported by previous research on fresh-cut papaya with a polysaccharide-based coating (Tabassum & Khan, 2020). Changes in total soluble solids due to starch hydrolysis that continued during fruit storage were due to the synthesis of sucrose and hexose in plant tissues, but this process became ineffective under low-temperature conditions. The inhibition of the increase in total dissolved solids indicates that the process of breaking down starch in the fruit is inhibited. Low O2 concentrations close to zero allow anaerobic respiration to occur, anaerobic respiration causes fruit to undergo fermentation so that the TSS value increases. In addition, papaya ripening is associated with increased activity of peroxidase and polyphenol peroxidase enzymes due to increased oxidative metabolism and ethylene accumulation in the fruit. The increase in TSS content in papaya fruit samples treated with edible coating with the addition of black tea extract which was higher than the control could be influenced by the edible coating layer that dissolved during the test.

3.2.5. pH

Table 5 shows, the highest change in pH value during the storage period in the edible coating sample of the papaya fruit with the addition

Table 6
Antioxidant activity and total carotenoids of papaya with minimal edible coating of black tea extract during storage.

Black Tea Extract Concentration	Storage (Day)	4	7	10
Antioxidant activity (%)				
Control	66.97 ±4.35 ^{bA}	70.93±5.19 ьв	71.16 ±2.21 ^{bC}	72±2.19 bC
0%	68.9±4.36 hA	70.5±6.08 ьв	70.45 ±2.93 ^{bC}	70.82 ±5.07 ^{bC}
5%	59.19 ±5.08 ^{aA}	$^{60.02\pm4.83}_{\scriptscriptstyle aB}$	69.87 ±2.65 aC	70.82 ±1.63 aC
10%	59.16±4.86	$\substack{63.09\pm3.84\\aB}$	70.99 ±1.6 ^{aC}	71.14±1.4 aC
15%	59.63±5.25 abA	$^{67.75\pm7.5}_{\scriptscriptstyle abB}$	70.48 ± 1.16^{abC}	72.63 ±2.33 abC
Total Carotenoid (µg/g)				
Control	9.51±1.47 bab	9.59±0.53 ьа	11.05 ± 1.16^{bAB}	12.71 ±1.67 bA
0%	8.97 ±0.73 ^{aAB}	9.49±1.43 aA	9.71±1 aAB	9.87±0.82
5%	9.6±0.12 abAB	$\begin{array}{l} 10.42 \\ \pm 1.51^{abA} \end{array}$	10.6 ±0.55 abAB	$10.91 \\ \pm 0.78~^{abA}$
10%	10.83±2.45 abAB	9.83±0.48 abA	9.65 ±1.08 abAB	9.35±0.29 abA
15%	$^{10.67\pm1.32}_{\scriptscriptstyle bAB}$	9.93±0.55 ьл	10.42 ± 1.9 bab	11.21 ±0.67 bA

of 15% black tea extract concentration. The pH value of the sample on day 1 was 5.86 and on day 10 was 5.31. While in the control sample the pH value on day 1 was 5.98 and on day 10 was 5.54. The results of statistical tests showed that there was no significant difference between the control treatment and papaya fruit with the edible coating (p>0.05). However, it can be seen in Table 5 that there was a significant change in the pH value during the storage period (p < 0.05). The storage, and lowering of pH during the storage period may be attributed to the increased microbial growth, followed by the production of organic acids (Mendy et al., 2019).

Coating provides a lower pH value than without coating, as well as cold storage. This shows that the application of an edible coating layer can inhibit the increase in the pH value caused by a decrease in the organic acid content during storage. Previous research showed that the addition of aloe vera with concentrations of 25% and 50% recorded the highest pH of 6.04 during the storage period. The increase in pH in this study could be a result of the coating of aloe vera gel which slows the ripening and aging of the fruit (Farina et al., 2020). The low pH of uncoated fruit could be due to a faster ripening process, where the low pH of uncoated fruit was caused by the production of acid from sugar metabolism at a faster rate as the fruit ripened (Tabassum & Khan, 2020). The steady increase in pH of coated fruit up to the last day of storage could be due to a decrease in fruit respiration rate.

3.2.6. Titratable acidity

The highest increase in titrable acidity (TAT) was papaya edible coating with the addition of 10% black tea extract of 0.36%, while the lowest was the control sample of 0.34% on the 10th day. The results of statistical tests showed that all treatments had no significant difference (p>0.05). Changes in TAT can be seen during the storage period on the 7th and 10th days there is a significant difference (p < 0.05) (Table 5). Total acid is closely related to the pH value, where an increase in total acid indicates a decrease in pH, so it can be seen that the acidity shown by an increase in acid in food can occur due to the breakdown of glucose into organic acid. The increase in the total value of titrated acid was also

due to the activity of microbial metabolites that produce acid. With the high activity of microbial metabolites with an increasing number of microbial populations, the results of microbial metabolites in the form of acids will be high (Tabassum & Khan, 2020). In addition, the increased total acid value was due to the production of organic acids that occurred in the respiration process at the tricarboxylic acid cycle stage. The organic acids produced in the tricarboxylic acid cycle include citric acid, fumaric acid, malic acid, and succinic acid. Before entering the tricarboxylic acid cycle, it must go through a carbohydrate degradation process, where this process produces glucose. Then glucose will be converted into pyruvic acid and enter the tricarboxylic acid cycle which produces water, carbon dioxide, and energy. Similar results have been reported by previous research thus implying that edible coatings might be effective in reducing the metabolic changes occurring in fruit or the rate of respiration thus delaying the consumption of organic acids (Tabassum & Khan, 2020).

3.2.7. Total carotenoids

Table 6 reports, that the highest total carotenoid content was in the control sample at 12,71 µg/g and the lowest was in the papaya fruit sample with an edible coating of 0% black tea extract by 9,35 µg/g on the 10th day. The results of statistical tests showed that there was a significant difference between the control sample and papaya fruit with an edible coating of 15% black tea extract with the addition of 0%, 5%, and 10% black tea extract (p < 0.005). In addition, there were significant differences during the 4th day of storage in all sample treatments (p < 0.05). Previous research also showed that the total carotenoids in papaya which were coated with the addition of aloe were 4.86 mg/100g FW for 9 days of storage (Mendy et al., 2019). As we know that carotenoids are natural organic pigment and lipophilic compounds found in several plants and are important parameters in that they impart attractive colors such as yellow, orange, and red (Ningrum & Schreiner, 2014). The content of carotenoid increase because of maturation and ripening that could be influenced by the storage. Uncoated fruits in this study have a rapid increase in carotenoids because the uncoated fruits ripen faster than coated fruits (Mendy et al., 2019).

3.2.8. Antioxidant activity

The increase in antioxidant activity of the black tea extract edible coating treatment sample was higher than the control sample during the storage period (Table 6). The highest increase was found in the addition of 15% black tea extract, namely on the 1st day of 59.26% and the 10th day of 72.62%. In the control sample, the antioxidant activity on day 1 was 66.97% and on day 10 it was 72%. Statistical test results showed that in the control sample and edible coating treatment with the addition of 15% there was a significant difference (p < 0.05), while during the storage period on the 7th and 10th days there was no significant difference (p>0, 05).

Tea polyphenols are known as strong antioxidants that can scavenge free radicals and reactive oxygen species (ROS) (Anggraini et al., 2016). Therefore, the antioxidant ability of the film can be increased by adding tea polyphenols. This change may also be associated with an increase in hydrogen and electron-donating ability after conjugation with tea polyphenols (Nenandis, 2008). The single electron transfer mechanism occurs when the antioxidant transfers one electron to the radical substrate which makes the antioxidant a radical intermediate or cationic radical. The resulting cationic radical antioxidant compounds are then deprotonated through interactions with water. The final step of this reaction is the same as that of the hydrogen atom transfer in the case of radical scavenging. The radical electrons and formal charge are initially present on the oxygen atom, but the electrons are thought to be delocalized and distributed throughout the aromatic ring. The ionization potential (IP) of antioxidant compounds is a parameter to predict the ability of antioxidants to scavenge free radicals through single electron transfer reactions. The larger the required ionization energy, the smaller the antioxidant molecule has the opportunity to donate electrons.

Table 7

Total Plate Count of papaya with minimal edible coating of black tea extract during storage.

Black Tea Extract	Storage (Da	y)		
Concentration	1	4	7	10
Total Plate Count (Log	CFU/mL)			
Control	6.05	6.81	7.17	7.38
	$\pm 0.32^{aA}$	$\pm 0.22^{aB}$	$\pm 0.16^{aC}$	$\pm 0.09^{aD}$
0%	3.69	5.9	6.46	6.97
	$\pm 3.21^{bA}$	$\pm 0.18^{\rm bB}$	$\pm 0.24^{bC}$	$\pm 0.17^{\rm bD}$
5%	1.67	6.03	6.75	7.37
	$\pm 2.89^{cA}$	$\pm 0.27^{cB}$	$\pm 0.09^{eC}$	$\pm 0.07^{cD}$
10%	5.8	6.58	6.97	7.32
	$\pm 0.48^{cA}$	$\pm 0.13^{cB}$	$\pm 0.17^{eC}$	$\pm 0.12^{\mathrm{cD}}$
15%	6.08	7.05	7.07	7.42
	$\pm 0.12^{dA}$	$\pm 0.17^{dB}$	$\pm 0.51^{dC}$	$\pm 0.15^{\rm dD}$

Previous research showed that the antioxidant activity of papaya fruit coated with the addition of aloe vera was 58.10% for 6 days of storage, while the 9th day was 65.21% (Mendy et al., 2019). In addition, it has been proven that chitosan has antioxidant properties, especially in terms of the ability to scavenge hydroxyl radicals (Salehi, 2020). Catalase activity prevents cells from fighting oxidative damage caused by stress by the breakdown of hydrogen peroxide into O_2 and water. Increased catalase activity was important to prevent cell damage while lower catalase activity indicated a weakened cell system to remove hydrogen peroxide.

3.3. Total plate count (TPC)

Microbiological observations on day 1 to day 7 in the control treatment showed a higher increase than the sample with edible coating treatment with the addition of black tea extract concentrations of 0%,

5%, and 10% (Table 7). In the control treatment, the number of microbes on day 1 was 6.04 Log CFU/mL and on day 10 was 7.38 Log CFU/mL. Treatment without the addition of black tea (0%) which counted on day 1 was 3.69 Log CFU/mL and on day 10 was 6.96 Log CFU/mL. Meanwhile, the addition of 5% black tea on day 1 was 1.67 Log CFU/mL and on day 10 was 7.36 Log CFU/mL. These data indicate that the microbial growth in the control sample was faster than in the edible coating treatment sample with the addition of black tea extract. The results of statistical tests showed that there was a significant difference between the treatment of black tea extract of 0%, 5%, 15%, and the control sample (p < 0.05). In addition, based on the statistical test of TPC testing on all samples during the storage period there was a significant difference (p < 0.05). So that the edible coating with black tea extract can have an influence on the number of microbes during the storage period.

The content of black tea extract contained tea polyphenols extracted from tea and contained catechins, flavones, anthocyanins, and phenolic acids, but catechins were the main component (Bansal et al., 2013). Catechins mainly contain epigallocatechin-3-gallate (EGCG), epigallocatechin (EGC), epicatechin-3-gallate (EGC), and epicatechin (EC) (Anggraini et al., 2016). Tea polyphenols in particular can coagulate structural proteins, bind deoxyribonucleic acid molecules, and destroy cell membranes and walls of microorganisms, thereby inhibiting the growth of microorganisms. Several previous studies have shown that chitosan can interact with negatively charged residues on the surface of microbial cells by an electrostatic attraction so that it flocculates and absorbs on the surface of microorganisms, then inhibits the physiological metabolism of microorganisms and finally inhibits the growth of microorganisms (Bansal et al., 2013). In addition, chitosan can affect microbial inhibition during storage (Badawy et al., 2017).

4. Conclusion

Edible coatings of composite gelatin-chitosan enriched with black tea extract exhibited a positive effect on fresh cut papaya shelf life

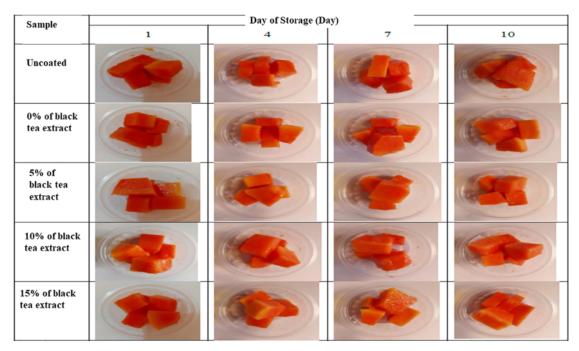


Fig. 2. Minimally processed papaya coated with chitosan-fish gelatin with different concetration of black tea extract and uncoted during refrigerated storage.

preserving its properties during a longer storage time than uncoated fruits. Edible coating with the addition of black tea extract could decrease weight loss and maintain minimallyy processed papaya firmness during storage. (p < 0.05) The edible coating also could influence the chemical properties of minimally processed papaya such as pH, total dissolved solids, total titrated acid, antioxidant activity, and total carotenoids. The edible coating can give a contribution a positive effect to protect the quality of minimallyy processed papaya showed especially to suppress microbial growth during storage. Therefore, this edible coating from gelatin-chitosan enriched with black tea extract can successfully increase papaya shelf life (Fig. 2).

Declaration of Competing Interest

The author(s) certify that they have No Conflict of Interest in the subject matter or materials discussed in this manuscript.

Data availability

No data was used for the research described in the article.

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