Research Article

Development of Whey-Chitosan Edible Films with Yellow Pumpkin Extract

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Abstract | Edible film made from whey protein isolate (WPI) and chitosan with adding yellow pumpkin extract has demonstrated excellent potential ability to make edible film that can extend food shelf life. The purpose of this study is to determine the characteristics of edible film. The method of this study used a completely random design with four treatments, namely without the addition of yellow pumpkin extract or control (T0), the addition of yellow pumpkin extract as much as 2.5% (T1), 5% (T2), and 7.5% (T3) and four replicates. The values obtained were collected through Microsoft Excel and processed using variety analysis (ANOVA); then, the data that showed significantly different results will be further tested with a Duncan test, while FTIR is analyzed qualitatively. The results of this study showed that the addition of yellow pumpkin extract had a highly significant effect ($P < 0.01$) increased opacity, WVTR, Permeance, color a*, b* color change, and yellowness index, but highly significant ($P < 0.01$) decreased swelling rate, L* color and whiteness index. Other properties like thickness, moisture content, tensile, elongation, young modulus, and WVP didn't show any significant results ($P > 0.05$). FTIR analysis shows that adding 7.5% of yellow pumpkin extract has a higher absorption band peak than other treatments. The addition of yellow pumpkin extract generally affects the characteristics of edible film.

Keywords | Edible film, Whey protein isolate, Chitosan, Yellow pumpkin extract

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INTRODUCTION

Packaging is one of the ways that can be used to protect food from environmental exposure. Food packaging is beneficial for slowing down product spoilage, delaying rapid food spoilage, and maintaining food quality (Kim *et al*., 2023). One of the most commonly used packaging is plastic. Plastic is a food product packaging widely used because it has superior properties, is transparent, waterproof, relatively cheap, and easy to get, but contributes to environmental pollution (Shih *et al*., 2011; Shit and Shah, 2014). As awareness of environmental issues, this has encouraged the use of packaging readily decomposed by the environment (Agustini *et al*., 2023). One of the packaging materials that can be used and has biodegradable properties is edible film.

Edible film is an ingredient that is an excellent barrier to moisture and oxygen and can store natural antioxidant compounds so that the oxidation process of food lipids and protein can be inhibited (Salehi, 2019). Edible film has been researched as an active functional ingredient that becomes an antioxidant and antimicrobial (Wongphan *et al*., 2022). Researchers have combined the preservation and packaging of biodegradable foods in edible packaging to retain moisture, prevent color fading, fat oxidation, undesirable odors, and increase shelf life and give good product characteristics (Umaraw *et al*., 2020). Edible film research continues to experience novelty, including raw materials used, manufacturing methods, and product application (Santoso *et al*., 2017). Therefore, it is necessary to design edible film making with materials that are easy to apply and have good edible film properties such as WPI and chitosan.

Chitosan (CH) is a type of carbohydrate and is a chitin-derived compound that can be found in fishery waste, namely shrimp shells and crab shells. Chitosan is often used as a base ingredient for packaging with antibacterial and antimicrobial properties (Perinelli *et al*., 2018). Chitosan has outstanding performance because it has properties like antifungal, antibacterial, biocompatible, and biodegradable (Ngo *et al*., 2015; Yuan *et al*., 2022). Whey proteins, specifically Whey Protein Isolate (WPI), are promising biopolymers and can create shiny, non-breakable edible film/coating with desired oxygen and oil barrier properties. WPI used as packaging has advantages such as easy decomposition, color that suits consumer desires, and a combination compounds like antioxidants and antimicrobials (Kandasamy *et al*., 2021; Jeon *et al*., 2023). It has been proven that the combination of proteins with polysaccharides can improve the emulsifying properties and stability of protein emulsions (Morales *et al*., 2021). Edible film will become more functional if they contain bioactive components to improve food quality. Using natural ingredients such as extract from plant can increase the effectiveness of packaging and prevent adverse effects such as oxidation of food (Ansarian *et al*., 2022). Plant extracts have many bioactives that can be obtained from bark, fruits, and vegetables (Nair *et al*., 2018).

Pumpkin (*Cucurbita moschata*) is a plant rich in bioactive compounds that have a positive impact due to their antioxidant abilities (Matić *et al*., 2024). Natural chemical compounds found in yellow pumpkin extracts such as vitamins, minerals, fiber, pectin, tocopherols, peptides, proteins, essential oil, amino acids, polysaccharides, phenolics, and carotenoids. The bioactives found in pumpkins have pharmacological and biological properties like antimicrobial, anticancer, antioxidant, antiinflammatory, antiaging, prebiotic, and cardioprotective properties. (Ezzat *et al*., 2022; Hussain *et al*., 2022). Pumpkins contain high β carotene, which causes the pumpkin to have an orange or yellow color (Halim *et al*., 2024). The potential of pumpkins to be developed in the active components of edible film is very large, but it is still not widely developed for the formation. The study purpose is findout edible film characteristics based on whey protein isolate and chitosan with the addition of yellow pumpkin extract, which helps extend the shelf life.

MATERIALS AND METHODS

The research material is edible film made from 2.5 g whey protein isolate (WPI 90 plain), 2.5 g chitosan (CH powder food grade), 1 mL glycerol (100% food grade), distilled water for WPI (100 mL), 2% acetic acid solution to dissolve chitosan (2 mL of acetic acid dissolved in 98 mL of distilled water) and materials for making yellow pumpkin extract (yellow pumpkin, citric acid and ethanol 70%).

The tools used include a glass beaker (Pyrex 250 mL), stirrer (duta nusantara type 79-1), filler pipette, magnetic stirrer, thermometer, timer, pan, label paper, grinder, microwave (Sharp), centrifuge, thermobath (THB-7 IWAKI), rotary evaporator (REN-1 IWAKI), electronic scale and black bottles for yellow pumpkin extract.

Extraction Method

The extraction method uses microwave-assisted extraction (MAE), which follows the procedure of (Hossain *et al*., 2024) with slight modifications: Yellow pumpkin (*Cucurbita moschata*) is cut into thin slices with a size of 5x5 cm^2 with a thickness of less than 0.5 cm. Then, it is dried in the sun (temperature 30 – 35°C). When it is dry, the yellow pumpkin was ground using a grinder. After smoothing, dissolve 1.5 g of citric acid in 100 mL of 70% ethanol in a beaker glass, then dissolve 10 g of yellow pumpkin flour in the earlier ethanol, then cover and let sit for 24 hours at room temperature. After that, 100 mL of solution is put in the microwave for 30 minutes (1 minute the microwave is on, 2 minutes the microwave is off). Once finished, the solution was centrifuged for 15 minutes at 2500 rpm. After that the solution is put into a rotary evaporator for 30 minutes at a temperature of 90°C. A total of 10 grams of pumpkin powder extracted with 70% ethanol produces 25 mL of thick extract.

The most dominant content in pumpkin extract is β carotene (Nurrahman and Astuti, 2022). Based on other studies (Qodri *et al*., 2024) 1 gram of pumpkin powder produces 43.995 µg / g β carotene with ethanol solvent. 10 grams of pumpkin powder that produces an extract of 25 mL of thick extract in this study is estimated to produce 439.95 µg / g β carotene or 0.43995 mg / g β carotene. The treatments in this study were P1 (addition of 5 mL extract estimated to contain 0.08799 mg/g β carotene), P2 (addition of 10 mL extract estimated to contain 0.17598 mg/g β carotene) and P3 (addition of 15 mL extract estimated to contain 0.26397 mg/g β carotene). The daily consumption range of β carotene is 1.5 - 1.8 mg/day (Bohm *et al*., 2021). Therefore, the concentration of pumpkin extract containing β carotene added to the edible film does not exceed this limit. In addition, in the pre-study, experiments and direct observations were carried out. The results were that the concentration gave the best results in the film. This is the basis for researchers to choose this concentration.

Edible Formation

The procedure for making edible film follows Qi *et al.* (2024) with slight modifications: Dissolve chitosan in 2% acetic acid solution. After that, it is stirred and heated using a stirrer for 30 minutes at a temperature of 80ºC. At the same time, dissolve WPI in distilled water and let it sit for 30 minutes after the chitosan solution is finished being homogenized. Then, the chitosan solution is left and continued by homogenizing the WPI solution for 10 minutes at a temperature of 70ºC. After completion, the WPI solution is put into the chitosan solution, then 1 mL of glycerol and the addition of yellow pumpkin extract according to the treatment and homogenized for 30 minutes at a temperature of 80ºC. After completion, the solution that has been made is cooled for ± 30 minutes, then poured into a mold as much as 30 mL. The criteria for a good edible film are not only environmentally friendly, but also have water, gas, and taste barrier properties as well as good mechanical properties (Venkatachalam *et al*., 2024). In addition, important properties in edible film are physical properties (Yang *et al*., 2024a) and color (Damayanti *et al*., 2023) in edible film.

Experimental Design

The research method used is a laboratory experiment using a complete randomized design of four treatments and four replicates modified from previous research. The treatment was given without adding yellow pumpkin extract as a control (T0), adding yellow pumpkin extract with a total of 0.25% (T1). 0.5%(T2) and 0.75%(T3). Treatment percentage from the total edible film solution made (a mixture of WPI and chitosan solution). This research was conducted at the Laboratory of Animal Product Technology, Faculty of Animal Science, University of Brawijaya. Fourier Transform Infrared (FTIR) Spectrofotometer testing is conducted in the central Mathematics and Natural Sciences laboratory at the State University of Malang.

Observed Variables

Thickness: The thickness measurement follows the procedure of Fang *et al.* (2025). It is done using a thickness gauge. The measurements were taken three times at different points, with each reading having an accuracy of up to 0.01 mm.

Moisture content: Moisture content measurement follows the procedure Sheikh *et al.* (2023): the edible film sample was cut to a size of 2 x 2 cm^2 and then weighed. Porcelain cups that have been dried in the oven are then weighed. The weight of the cup is calculated as W0 and the weight of the sample and the cup is calculated as W1. Then, the sample is put into a porcelain cup and baked at 105ºC for 24 hours. The sample is then put in a desiccator, weighed, and calculated as W2. After that, the moisture content is calculated by the formula:

$$Moisture\ Content = \frac{W1 - W2}{W1 - W0} \times 100$$

Swelling rate: The method used in measuring the swelling rate follows Bhatia *et al.* (2024). The edible film sample is cut into 2 x 2 cm^2 sizes and then weighed (W1), then the sample is soaked in distilled water for 2 minutes in a porcelain cup. After that, the edible film is then wiped using a tissue and weighed (W2). The swelling rate value is calculated using the formula:

$$Swelling\ rate = \frac{W2 - W1}{W2} \times 100\%$$

Note: The 2 minutes time selection was based on previous research (Bhatia *et al*., 2024; Erdem *et al*., 2019). In addition, this observation time is sufficient for the development of edible film at the beginning, the time is fast and shows the hydrocolloid properties of edible film.

Water solubility: The solubility test procedure follows the method carried out by Gumus *et al.* (2024): the edible film sample was cut into 2 x 2 cm^2 and then dried in an oven at 105ºC until constant weight, then left in a desiccant and weighed (W1). Then, an aluminum cup is prepared and filled with equates until it fills half and leaves for 24 hours at room temperature. After that, the samples are filtered and baked at 105ºC until their weight is constant. Then, the sample is weighed as (W2) and calculated by the formula:

$$Water\ solubility = \frac{W1 - W2}{W1} \times 100$$

Water barrier properties: The measurement water barrier properties based on ASTM E96/E96M-16 (Rawat and Saini, 2024) with modification: the sample was cut in a circle with a diameter of 4.5 cm. The aid uses a tube container with a circle diameter of 4.3 cm and a height of 4 cm, the lid is perforated so that water transmission can occur. The tube-shaped aid is filled with as much as 3/4 of its volume of distilled water. The circular sample is placed on the top of the aid and then closed using the lid of the tool that has been punched. The sample is weighed and determined to be the initial weight of the sample. The sample is placed in a desiccant (the humidity of the desiccator is 0% (R0), and the humidity in the sample is estimated to be 100%(R1)). The sample is weighed after 24 hours to determine the final weight. The following formulas calculate water barrier properties:

Water Vapor Transmission Rate

$$WVTR = \frac{\Delta W \times \Delta t}{Area}$$

Where:
ΔW/Δt = Weight loss of edible film/time change

Permeance

$$Permeance = \frac{WVTR}{\Delta P}$$

$$\Delta P = \frac{R1 - R0}{100} \times Pvap.sat$$

Where:
R1 = Moisture in the edible film system
R0= Moisture system outside edible film (desiccator)
Psat. vap= Water-saturated steam pressure

Water Vapor Permeability

$$WVP = Permeance \times WVTR$$

Mechanical Properties

These measurements are based on ASTM D828–97 2016 (Niu *et al*., 2024) with a modification: The sample was cut into squares of 1 x 6 cm^2 and then formed according to a pattern that followed the provisions of the Physics Laboratory University of Brawijaya. The sample was analyzed in thickness using a thickness gauge with three repetitions and then recorded. The sample was analyzed using tensile strength, and after the thickness was known, the elongation value was recorded. The final result contains data to calculate the variables of the mechanical properties of the sample; here is the formula (Niu *et al*., 2024):

$$Tensile\ strength\ (TS) = \frac{Fmax}{w \times d}$$

$$Elongation\ at\ Break\ (EAB) = \frac{L1}{L0} \times 100$$

$$Young\ Modulus\ (YM) = \frac{Tensile\ Strength}{Elongation\ at\ Break}$$

Where:
Fmax= Max force when sample breaks
w= Sample width (5 mm)
d= Thickness of edible film
L1= Sample extension
L0= Initial length

Optical Properties and Color Measurement

Opacity: Measurement of opacity follows the Biratu *et al.* (2024): The edible film sample was cut to 1 x 3 cm^2. The sample was then fed into a quartz cuvette and analyzed using a single-beam UV-VIS spectrophotometer with a wavelength of 600 nm. The absorbance value is recorded and calculated with the following formula:

$$Opacity = \frac{A600}{d}$$

Where:
A600= Absorbance at 600 nm
d= Edible film thickness (in millimeters)

Color measurement: Color measurements are based on Erdem and Kaya (2022): The sample was cut with a size of 5 x 5 cm^2. The samples were analyzed using Minolta CR-10 from the Animal Products Laboratory for three replicates. The results were obtained from L^*, a^*, and b^* colors, and then the color difference (ΔE), whiteness index (WI), and yellowness index (YI) were calculated. Here is the formula (Erdem and Kaya, 2022):

$$\Delta E = (\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2})^{1/2}$$

$$WI = 100 - [\,(100 - L^*)^2 + (a^*)^2 + (b^*)^2\,]^{1/2}$$

$$YI = \frac{142 \cdot 86 \times \Delta b*}{\Delta L*}$$

Fourier transform infrared (FTIR) spectroscopy: FTIR follows the procedure described by (Tan *et al*., 2024). FTIR spectroscopy (IRTracer, SHIMADZU, Japan) was performed in the spectral range of 4000–400 cm− 1.

Data Analysis

The data obtained is tabulated using Microsoft Excel. The data was statistically analyzed by calculation using analysis of variance (ANOVA) single factor according to the method used, Complete Random Design. If results are significant or highly significant between treatments, continue with the Duncan Multiple Range Test (DMRT) (Hashemi *et al*., 2021).

RESULTS AND DISCUSSION

Thickness

Thickness is a character that needs to be considered in edible packers with a maximum standard of less than 0,25 mm (Japanese Industrial Standard, 2019). Data in this study show that the thickness value of edible film ranging from 0.086 - 0.131 mm (Table 1). The addition of yellow pumpkin extract showed a significant value ($P < 0.05$). This increase in thickness is due to the total solids from the yellow pumpkin extract on the film so that it can cause pores.

Gao *et al.* (2024) found that the thickness increases with addition of anthocyanins. Another factor that increases the thickness is the cohesion properties of the edible film. Cohesion causes inner interactions in the same material, especially if the particle size is smaller (Lin *et al.*, 2024). The peak of the edible film improvement was in T2 and fell in T3. This can be caused by the uneven distribution of the total solids of yellow pumpkin extract. The different thickness of each edible film can be affected by the different composition of the solution, because the thick solution tends to produce a thicker edible film (Rawat and Saini, 2024). Thickness affects the edible film for the product it is coated with. This variable is adjusted to the packaged product. The amount of water content in edible film affects the thickness, that is the greater the water content in the material, it can increase the thickness with the same surface area (Coniwati *et al.*, 2014).

Table 1: Physical and optical variables on WPI-CH film with the addition of yellow pumpkin extract.

Films	T (mm)	MC (%)	SR (%)	WS (%)	Opacity (mm^{-1})
T0	0.086 ± 0.011[a]	7.35 ± 0,687	54.535 ± 6.39[b]	33.40 ± 8.81	3.51 ± 0.61[a]
T1	0.110 ± 0.024[ab]	7.55 ± 0,797	41.997 ± 7.15[ab]	38.54 ± 6.04	3.23 ± 0.84[a]
T2	0.131 ± 0.012[b]	7.80 ± 0,368	28.605 ± 4.18[a]	38.66 ± 6.94	7.12 ± 0.95[b]
T3	0.106 ± 0.018[ab]	7.36 ± 0,618	28.942 ± 9.92[a]	41.43 ± 8.52	11.02 ± 1.85[c]

Where: T (Thickness); MC (Moisture content); SR (Swelling rate); WS (Water solubility).

Moisture Content

The results of this study showed that the water content ranged from 7.35-7.80% (Table 1). The water content increased with the addition of pumpkin extract. This can be caused by the addition of pumpkin extract as a liquid that can make water fill the polymer chain (Bhatia *et al.*, 2024). Another study by Mirsharifi *et al.* (2023), the water content of edible film made from almond gum/polyvinyl alcohol/chitosan with thyme essential oil had the highest value of 33.15%, while the control water content had a value of 7.95%. However, in this study, the water content value increased from T1 (7.35%) to T2 (7.55%), while T3 (7.36%) decreased, and the difference in water content was not significant (P>0.05). This happens because the increased thickness value with pumpkin extract can increase the water content in the edible film. The water content value has the same pattern as the thickness of the edible film in this study. In addition, the water content value is influenced by matrix stability caused by polyphenol compounds that interact with the protein polypeptide chain. The interaction of these compounds can maintain the stability of noncovalent bonds of protein and phenol complexes (Manzoor *et al.*, 2024). The low water content in this study provides advantages as primary packaging because it does not contribute water to the product which causes a decrease in product quality (Rusli *et al.*, 2017).

Swelling Rate

The water resistance properties of edible film are determined using the swelling test, which is the percentage of swelling of the edible film by water. A high value of swelling show that edible film absorbs high water (Susilowati and Lestari, 2019). The swelling value tended to be highly significant (P < 0.01) with swelling rate value ranging from 28.60 to 54.53%. This indicates that edible film with the addition of pumpkin extract reduces the level of swelling. Yellow pumpkin is a fruit rich in carotenoids and polyphenols (Hussain *et al.*, 2022). Carotenoids are lipophilic compounds (Mussagy *et al.*, 2022) that will interact with the hydrophobic group (Gomes *et al.*, 2021). The lower value show that good edible film produced. However, every treatment of the edible film has good stability, the edible film remains intact when submerged in water (Yang *et al.*, 2024b). A short soaking of the edible film (2 minutes) may reduce the film's swelling value. Each edible film has a different hydration time depending on the primary polymer used in the film. The swelling rate value in this study has different variations, the control is not suitable as a primary coating because it can absorb water content in food while other treatments can be applied as a food primary coating because of its low water absorption ability.

Water Solubility

A study on the water solubility of edible film was carried out to determine the ability of packaging to degrade in water. Water solubility value is related to the permeability of water vapor. This can be seen from hydrophilic groups that can interact with edible films, especially interactions with H_2O (Agustini *et al.*, 2023). The results of this study show that WPI-CH edible film with yellow pumpkin extract has a value ranging from 33.40 to 41.43% (Table 1). The addition of yellow pumpkin extract increases the film's solubility in the solvent. The content of yellow pumpkin β carotene, α tocopherol, vitamin C, and vitamin A (Kim *et al.*, 2016). Some substances can be soluble in polar solvents and non-polar solvents. Vitamin C is soluble in water, but vitamin A is insoluble in water and fat-soluble. Carotene is a compound that is fat-soluble but insoluble in water (Mussagy *et al.*, 2022). Fructans, pectins, and arabinoxylan are fibers from yellow pumpkins that increase water solubility because they can bind water (Halim *et al.*, 2024). Solutes in vegetable oils (e.g. sunflower oil, olive oil, and palm), like tocotrienols and tocopherols have polar groups such as cromanol rings, these compounds can be soluble in polar solvents (Gonzalez–Diaz and García–Núñez, 2023).

This water-soluble compound can increase solubility with the addition of yellow pumpkin extract. However, this variable did not have significant value (P > 0.05). Application of edible film to foods that have a high moisture content is preferable if it has a low solubility value (Sahraee *et al.*, 2017; Liu *et al.*, 2023). It can be applied to foods with high water content. Table 2.

Table 2: Variable water barrier properties on WPI-CH edible film with yellow pumpkin extract.

Films	WVTR (g/m²s). 10^{-10}	P (g/m²s. Pa). 10^{-13}	WVP (g/m²s. Pa). 10^{-15}
T0	3.78 ± 0.16^a	1.19 ± 0.05^a	12.18 ± 3.26
T1	4.07 ± 0.02^b	1.28 ± 0.01^b	13.95 ± 2.14
T2	4.11 ± 0.07^b	1.29 ± 0.02^b	16.98 ± 4.84
T3	4.31 ± 0.14^b	1.36 ± 0.04^b	14.02 ± 2.57

Where: WVTR (Water Vapor Transmission Rate); P (Permeance); WVP (Water vapor permeability).

Water Barrier Properties

The properties of water barriers are seen in WVTR, Permeance, and WVP. The WVTR values range from 3.78 to 4.31 (g/m²s). 10^{-10}. The permeance value in this study ranged from 1.19 to 1.36 (g/m²s. Pa). 10^{-13}. The WVP value in this study ranged from 12.18 to 16.98 (g/m²s. Pa). 10^{-15}. The addition of yellow pumpkin extract contributed to increasing the WVTR value and edible film permeance, and the value is highly significant (P < 0.01). This can occur because the addition of pumpkin extract increases water diffusion in the film. Diffusion occurs during water permeation through edible film, where vapor passes through the matrix from the high to the lower concentration (Silva *et al.*, 2018). Water vapor transmission rate and permeance are also caused by matrix surfaces and pores in edible film (Srinivasa *et al.*, 2007; Erdem *et al.*, 2019). The high thickness of the edible film with the addition of yellow pumpkin extract is suspected to have a large pore content compared to the control. WVP value shows that treatment contributed to increasing its value but did not significantly (P > 0.05). The value of WVP indicates how much water can pass through the edible film in a certain period (Gumus *et al.*, 2024). Lower WVP indicates reduced chain mobility in edible film due to the presence of crosslinking, as seen in edible film made of cellulose with Ca^{2+} ions (Xu *et al.*, 2016; Paudel *et al.*, 2023; Sigdel and Janaswamy, 2024). WVP edible film is affected by solubility, edible film surface thermodynamics, and diffusivity (Jeon *et al.*, 2023). The control in this study had a good water barrier, but the value of the addition of 2.5% yellow pumpkin extract had a value close to the control and had a good ability value in blocking water. Good water barrier properties in edible film are important as packaging to protect the food in the package (Dewi *et al.*, 2021).

Optical Properties and Color Measurement

Light barrier properties are important in edible films to protect light-sensitive foods such as fresh meat (Farhan and Hani, 2020). The opacity value can be seen in Table 1. The opacity value of this study ranged from 3.23 to 11.02 mm-1 (P <0.01). The opacity value in the control and the addition of 2.5% pumpkin extract was lower than the opacity value with the addition of extract above 2.5%. A low opacity value indicates that the film is brighter and light can penetrate the film effectively (Aprilliani *et al.*, 2019). The varying opacity values indicate differences in the thickness of the edible film and the constituent compounds in the edible film matrix. Low opacity values are preferred because they are transparent, but high opacity values can also be preferred in certain products, for example, products that are sensitive to light (Biratu *et al.*, 2024).

Table 3: Variable color measurement on WPI-CH edible film with the addition of yellow pumpkin extract.

Films	L*	a*	b*	ΔE	WI	YI
T0	86.01 ± 2.30^a	0.16 ± 1.02^a	23.18 ± 6.55^a	23.35 ± 6.98^a	72.88 ± 6.81^a	38.74 ± 12.08^a
T1	83.98 ± 2.69^a	16.54 ± 2.52^b	28.32 ± 7.24^a	29.32 ± 8.04^a	67.15 ± 7.88^a	48.51 ± 14.16^a
T2	72.74 ± 5.07^b	52.24 ± 3.51^d	46.44 ± 7.75^b	51.85 ± 9.74^b	44.57 ± 9.80^b	92.28 ± 20.90^b
T3	73.59 ± 4.20^b	41.66 ± 3.39^c	47.16 ± 6.01^b	51.53 ± 7.62^b	44.90 ± 7.68^b	92.22 ± 16.10^b

Where: L* (lightness); a*+ (Red); a*- (Green); b*+ (Yellow); b*- (Blue); ΔE (Different color); WI (Whiteness index); YI (Yellowness index).

Color measurements are essential in edible films because these properties will affect consumer acceptance of food products (Kevij *et al.*, 2020). The color measurement value can be seen in Table 3. The L* value in this study ranged from 72.74 to 86.01, and the value was highly significant (P < 0.01). An increase in yellow pumpkin extract may decrease the L* value, but the addition of 2.5% yellow pumpkin extract does not differ significantly from the control. Another study with an increase in walnut oil content from 0.5 to 1% led to a slight decrease in L* values ranging from 89.8 to 90.8 (Galus and Kadzińska, 2016). a* value ranged from 0.16 to 52.24 with the addition of yellow pumpkin extract, and the value was highly significant (P < 0.01). The value of b* ranged from 23.18 to 47.16, which was highly significant (P < 0.01). The addition of yellow pumpkin extract influenced the b value, and the effect was pronounced when adding 5% and 7.5% yellow pumpkin extract. Another study observed a positive increase in the b* value (1.99 to 2.51) due to increased cinnamon bark oil (1% to 3%) matrix of edible film with chitosan and the results of the study sunflower and cinnamon can be attributed to the natural yellow color (Ma *et al.*, 2015; Erdem and Kaya, 2022). ΔE has a value ranging from 23.35 – 47.16, which is highly significant ($P < 0.01$). The control ΔE value was not significantly different in the treatment with adding 2.5% yellow pumpkin extract but was very noticeable when adding 5% and 7.5% yellow pumpkin extract. The color that tends to be yellow, with the addition of yellow pumpkin extract listed in Table 3, will affect the yellowness index of the film. The yellowish index value in this study ranged from 38.74 to 92.28, and the value was highly significant ($P < 0.01$). Meanwhile, the whiteness index will have the opposite value of the yellowish index, with values ranging from 44.57 to 72.88 and highly significant ($P < 0.01$). Yellow pumpkin extract exerts a noticeable effect on the color change of the film. Yellow pumpkin is rich in β-carotene which has an orange color (İzli *et al*., 2022). In addition, the thickness of the edible film also contributes to the color measurement results of the edible film.

Table 4: Variable mechanical properties on WPI-CH edible film with the addition of yellow pumpkin extract.

Films	TS (MPa)	EAB (%)	YM (MPa)
T0	15.19 ± 1.95	21.67 ± 8.05	0.78 ± 0.36
T1	16.59 ± 4.54	15.97 ± 2.77	0.92 ± 0.64
T2	14.34 ± 4.36	20.25 ± 4.80	0.73 ± 0.30
T3	12.75 ± 1.86	21.17 ± 9.32	0.70 ± 0.35

Where: TS (Tensile strength), EAB (Elongation at break), and YM (Young modulus).

Mechanical Properties

The film's tensile strength, elongation at break, and young modulus or elasticity are indispensable for food packaging materials because these properties can protect food products, so good mechanical properties are needed (Ahmadzadeh *et al*., 2023). The tensile (TS) value in this study ranged from 12.75 to 16.59 MPa. The elongation value in this study ranged from 15.97 to 21.67 %. The value of the young modulus or stiffness ranges from 0.70 to 0.92 MPa. The tensile strength related to the cohesion force between the chain and elongation can show the film's ability to stretch (Galus and Lenart, 2013; Tavares *et al*., 2021). Tensile strength value in this study showed that adding 2.5% yellow pumpkin extract could increase tensile strength but decrease the elongation of the film. However, adding 5% and 7.5% yellow pumpkin extract decreased tensile strength ($P > 0.05$). The decrease in tensile and increase in EAB value occurred after added essential oil edible film (Bhatia *et al*., 2024). This effected can be caused by the structure of the tissue formed in the edible film so that it can affect the hardness of the edible film (Zheng *et al*., 2024). The value of mechanical properties is influenced by phenolic compounds and phenolic interactions with the materials that make up the edible film so that it can cause a plasticization effect that affects the mechanical properties (Nie *et al*., 2015; Jiang *et al*., 2022). The elasticity value in all treatments was categorized as less elastic ($P > 0.05$), but a higher value was seen in the addition of 2.5% yellow pumpkin extract. The low elasticity value is < 20 MPa, while the high elasticity value is 100 – 110 Mpa (Cortés-Rodríguez *et al*., 2020). The data shows that the mechanical properties of T2 outperform T3, this could be because the edible film of T2 is thicker than that of T3. Good edible film for food packaging is strong and elastic (Ningrum *et al*., 2021). Table 4.

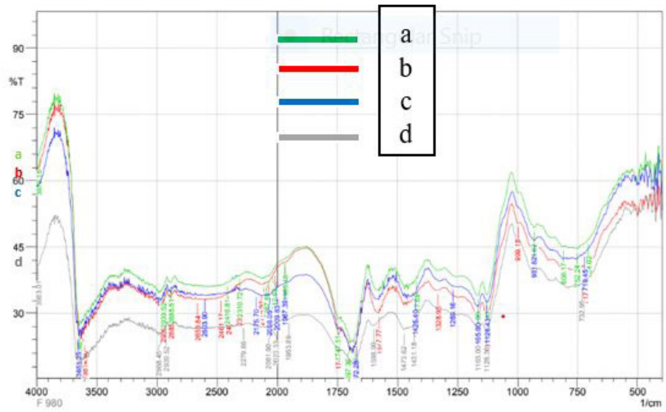

Figure 1: FTIR. FTIR bands are symbolized as a, b, c and d. where: (a) Without the addition of yellow pumpkin extract (T0), (b) Addition of 2.5% yellow pumpkin extract (T1), (c) Addition of 5% yellow pumpkin extract (T2), and (d) Addition of 7.5% yellow pumpkin extract (T3).

Fourier Transform Infrared (FTIR) Spectroscopy

FTIR analysis was used to determine the chemical interactions and functional groups that occur in edible film. In the food industry, this controls product quality (Indrianingsih *et al*., 2024). In this study, the FTIR results are shown in Figure 1. The results showed that at 3600 cm^{-1} waves, it is estimated that there are -OH and -NH groups with a range of 3438.17 cm^{-1} to 3631.96 cm^{-1}. This is likely an interaction amine group in chitosan and the OH group in cellulose. In another study, the sharp peak of the –OH group at 3631.96 cm^{-1} indicated the presence of free –OH group in microcrystalline cellulose (Yasmeen *et al*., 2016). At 2900 cm^{-1} wave, there is alkane group. This can be indicated by the content of β carotene ($C_{40}H_{56}$) from yellow pumpkin extract (Gowtham *et al*., 2022). At 1700 cm^{-1} wave, there is a C=O group in the form of an ester from yellow pumpkin extract (Gowtham *et al*., 2022). At 1628 cm^{-1} corresponds to the amide II band (vibration –C = O), and at 1535 cm^{-1} due to the amide II band (stretch –C = O and C–N) (Tavares *et al*., 2021). In this wave range, Schiff formation (Schiff bases are defined as chemical compounds (imines) bearing a hydrocarbyl group on the nitrogen atom (Raczuk *et al*., 2022) occurs between chitosan and WPI (Muley and Singhal, 2020). At waves ranging from 1200 – 1300 cm-1, N=O, S=O, C-N, N-O, P=O from yellow pumpkin extract (Muley and Singhal, 2020). Other studies have found that the crest is located at about 1240 – 1230 cm^{-1}, possibly due to the bend of the amida III (Erdem and Kaya, 2022). The spectral band in the amide region I, range from 1633 cm - 1332 cm^{-1} can be credited to the interaction of phenolic compounds and flavonoids from proteins (Jridi *et al*., 2020; Das *et al*., 2024).

Figure 1 show that the gray band has a higher peak compared to other treatments. This can happen because increase the concentration of yellow pumpkin extract can increase the number of groups derived from yellow pumpkin and interactions between molecules. Molecular interaction can strengthen the molecular integrity of edible film structure and improve edible film resistance (Farhan and Hani, 2020). In another study (Thakur *et al*., 2024) explained the results of their research that an increase in the concentration of chitosan lactate (ChL) by up to 1%(b/v) increased the intensity and dilatation of FTIR peaks, indicating higher intermolecular interactions compared to other treatments. Yellow pumpkin extract with different concentration gives different appearance or graphic results, such as the hydrophobic properties of the ester and β carotene groups and the addition of orange color to the packaging.

CONCLUSIONS AND RECOMMENDATIONS

The addition of yellow pumpkin extract to WPI-chitosan-based edible film affected the swelling rate, WVTR, Permeance, opacity, and Lab color. The FTIR results in the edible film, with an additional 7.5% yellow pumpkin extract in edible film increase in the band peak, indicate a high intermolecular interaction between β carotene in yellow pumpkin extract with WPI and chitosan. The addition of yellow pumpkin extract with different concentrations also provides different value of edible film. The addition of 2.5% yellow pumpkin extract in edible film tended to have a close physical character to the control, while 5% and 7.5% had similar physical characteristics. Edible film with the addition of yellow pumpkin extract can be further developed into a smart film because the content of carotene β has an orange color which can be used as a color indicator.

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NOVELTY STATEMENTS

This research is novel in developing raw materials.

AUTHOR'S CONTRIBUTIONS

All authors contributed equally.

Conflict of Interest

There is no conflict of interest from author.

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