

Autoclaving–Cooling Modified Purple Yam (*Dioscorea alata* L.) Starch for the Development of Edible Films

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Abstract

This study aimed to analyze the effect of purple yam (*Dioscorea alata* L.) starch modification through repeated autoclaving–cooling cycles and different starch concentrations on the characteristics of edible films, as well as to determine the best treatment combination. A factorial Completely Randomized Design (CRD) was applied with two factors, namely starch modification (native, one cycle, and two cycles) and starch concentration (5%, 6%, and 7%), with three replications, resulting in 27 experimental units. The results showed that starch yield significantly increased from 20.42% (native) to 90.13% (one cycle) and 98.16% (two cycles). Starch pH increased from 5.80 to 6.81–6.89, moisture content decreased from 8.72% to 7.26%, and crude fiber rose from 5.22% to 6.56%. SEM analysis revealed progressive granule disruption, producing edible films with denser, more homogeneous, and crack-free structures. Starch color parameters indicated decreased lightness (L^*) from 67.49 to 65.59, increased a^* from 5.95 to 6.39, and increased b^* from 13.20 to 14.35. In contrast, edible film color varied with L^* values of 71.96–77.47, a^* 3.56–5.17, and b^* 14.78–18.91, influenced by the interaction between starch concentration and modification cycles. Anthocyanin content increased both in starch (from 3.17 to 5.82–5.86 mg/g) and edible films (from 8.07 to 11.10–11.12 mg/g). Functional properties of the edible films showed stable thickness (0.20–0.24 mm), variable transparency and solubility, reduced water vapor transmission rate (WVTR) down to 3.69 g/m²·day, and improved tensile strength up to 292.33 g. In addition, crude fiber content in edible films increased up to 4.70%, with relatively stable pH ranging from 5.72 to 6.17. The novelty of this study lies in the application of repeated autoclaving–cooling on purple yam starch, which not only enhanced the technological properties of edible films (thickness, transparency, mechanical strength, and barrier performance) but also increased anthocyanin content, resulting in dual-function edible films as eco-friendly biodegradable packaging and a natural antioxidant source.

Introduction

Purple yam tuber plants (*Dioscorea* spp.) produce tubers that are classified as aerial tubers and tubers found in the soil, this plant is a monocotyledonous annual plant, grows creeping with a winding direction to the right, the stem length reaches 10 meters, is not thorny but some are spotted at the base, the stem is four-angled with wings, green or purplish in color, often there are tubers in the leaf axils and has a single leaf, curved leaf veins, with seven to nine leaf veins, green or purplish in color, oval leaf blades with a heart-shaped base and a long tapering tip, fibrous root system. Yam plants (*Dioscorea* spp.) have spike-shaped flowers, male flowers are densely spiked, female flowers are sparsely spiked, inflorescence occurs in May-June, with flat, rounded wings around them. Underground tubers have various shapes and sizes, (Lathifah et al., 2024)

One potential source of starch that can be utilized is purple yam tuber (*Dioscorea alata* L.). This plant is rich in starch and contains anthocyanin pigments that act as natural antioxidants (Nassour et al., 2020). This potential makes purple yam not only a local food ingredient, but also a candidate for raw material for edible films with higher functional value. Unfortunately, natural purple yam starch has weaknesses in

the form of low yield, high water content, and less stable film mechanical properties. Therefore, the application of physical modification through repeated autoclaving-cooling is expected to improve the quality of the starch while strengthening the characteristics of the resulting edible film.

Various starch modification methods have been developed to overcome these limitations, one of which is physical modification using the autoclaving-cooling method. Autoclaving-cooling treatment causes gelatinization of starch granules at high temperatures and pressures, followed by retrogradation during cooling, resulting in changes in crystalline structure, increased resistant starch, and improved functional properties (D. N. Faridah et al., 2022; Oktaviani et al., 2023). This modification not only improves the physicochemical quality of starch but also has the potential to protect and increase the availability of bioactive compounds such as anthocyanins.

Previous studies have shown that *autoclaving-cooling modification* of various types of starch can increase stability, improve physical structure, and enrich functional components (Oktaviani et al., 2023). Furthermore, this treatment has been shown to affect water content, pH, fiber, and bioactive pigments, which in turn determine the mechanical and barrier properties of edible films (K. Wang et al., 2017). However, studies on the application of the repeated *autoclaving-cooling method* to purple yam starch, especially for edible film applications, are still limited. This study aims to analyze the effect of modifying purple yam starch (*Dioscorea alata L.*) through the *autoclaving-cooling method* and variations in concentration on the characteristics of *edible films*, and to determine the best treatment combination.

Research methodology

Materials and tools

The materials used are purple yam tubers, water, distilled water, the tools used in this study are analytical scales, sieves, blenders, 200 mesh sieves, baking pans, knives, basins, electric ovens, cutting boards, beakers, measuring cups, stirring rods, magnetic stirrers, plastic clips, hot plates, petri dishes and analytical tools in the form of thermometers, test tubes, screw tubes, desiccators, *texture analyzers*, filter paper, *scanning electron microscopes* (SEM) and spectrophotometers, Color Readers.

Purple Yam Starch Processing with Autoclaving–Cooling

Uwii starch was suspended in 20% water (treated with a 20% water content adjustment), and stored in a refrigerator at 4°C for 12 hours to evenly distribute the water in the starch. Then, it was heated using an autoclave at 121°C for 15 minutes. The starch was then immediately cooled at room temperature for 1 hour to prevent further gelatinization. Next, the starch was retrograded by cooling at 4°C for 24 hours. For the autoclaving-cooling treatment, 2 cycles of heating with an autoclave and cooling at 4°C were repeated once more. After that, it was dried in an oven at 50°C for 4 hours. The dried starch was sieved using a 200 mesh sieve, and the starch was analyzed to determine its physicochemical properties. (Lehmann et al., 2002).

Making Edible Film

Weigh Uwi starch according to concentration is 5% 6% and 7% or 5 g, 6 g, and 7 g, to in 100 ml distilled water, stirred until homogeneous. Solution starch then heated while stirring until gelatinization at a temperature of 80°C – 90°C for ± 6 minutes. Add glycerol with a concentration of 4% of the starch weight. Stir for ± 5 minutes minute until homogeneous. After That pour to mold cup petri as much as 25 ml and dry in an oven at 50°C for 6 hours, after drying, store in a closed container (Piñeros-Hernandez et al., 2017).

The experimental design used was a Completely Randomized Design (CRD) in the form of a factorial experiment, with two factors: starch concentration and *autoclaving cycle*. Each treatment was repeated three times, resulting in 27 samples:

Factor I: Starch concentration

I1: starch concentration 5%

I2: starch concentration 6%

I3: starch concentration 7%

Factor t: *Autoclaving cycle*

t0: natural starch (without cycle)

t1: one cycle modified starch

t2: Modified starch two cycles

Research Parameters

a. Modified Yam Starch Yield

Yield is the ratio of the dry weight of the product produced to the weight of the raw materials used (Moorthy, 2002). Modified yam starch was calculated to determine the yield of modified starch using the following formula:

$$Rendemen (\%) = \frac{a}{b} \times 100$$

b. pH measurement

Weigh 1 g of starch and put it into a test tube. Then add 20 mL of distilled water and vortex for 5 minutes until homogeneous, then put the solution into a beaker glass. pH measurement using a pH meter, where

before use the pH meter is turned on and let it stand until stable (15–30 minutes). Rinse the electrode with distilled water, dry it with tissue paper. Dip the electrode into the starch solution and measure the pH (Moorthy, 2002).

c. Level Water

The empty cup was oven-dried for 15 minutes, cooled in a desiccator, and then weighed. A 3 g starch sample was placed in the weighed cup and then dried in an oven at 100–105°C for 1 hour. The cup containing the sample was removed. to desiccator, cooled, and weighed. Drying is carried out again until a constant weight is obtained. The water content is calculated based on the weight loss, namely the difference between the initial weight (W1) and the final weight (W2). The determination of water content is based on the equation (Ben-Gigirey et al., 2012)

$$K a_{bk} = \frac{W_a}{W_k} \times 100 \% = \frac{W_t - W_k}{W_t - W_a} \times 100 \%$$

d. Color

Edible film color analysis was performed using a CS-10 digital colorimeter. The color measurements on the film samples were first calibrated with a black standard, then the target readings were determined. The colors were L* (0 = black- 100 = white), a* (-60 = green, + 60 = red), and b* (-60 = blue, + 60 = yellow) (Jridi et al., 2013).

e. Fiber Content

Whatman No. 41 filter paper was dried in an oven at 105 °C for 1 hour and weighed its initial weight (Z). Weighed 2 g of starch sample carefully and put it into a 250 ml Erlenmeyer flask. Add 50 ml of 0.255 N H2SO4 and heat in a condenser for 30 minutes, then quickly add 50 ml of 0.313 N NaOH, and heat again for 30 minutes. The liquid was then filtered using filter paper of known weight. The filtered sample was rinsed using hot distilled water, if the filtered water was clear, add 15 ml of alcohol after that, oven the filter paper for 3 hours, calculated using the equation (Ben-Gigirey et al., 2012).

$$\text{Serat kasar } \% = \left(\frac{\text{berat akhir} - \text{berat kertas saring}}{\text{berat sampel}} \right) \times 100$$

f. Morphology of Starch Granules

Natural and modified yam starch were tested using a Scanning Electron Microscope (SEM) (model JEOL JSM 6510 LA) which had previously been dispersed using alcohol. The sample was placed on an aluminum stab using double-sided adhesive tape and coated with gold powder to avoid charging under the electron beam after the alcohol evaporated, the starch granules were observed at a magnification of

1000×. Measurement of starch granules used the ImageJ application version 1.5.2 with a calibration scale found in the SEM image, then the measurement of starch granules was carried out by measuring the length of the granules, (Gunawan et al., 2023)

g. Anthocyanin Content

Analysis of anthocyanin levels was carried out using the differential pH method referring to with modifications (Lee et al., 2005), sample extraction was carried out using 70% ethanol which was left with HCl IN (85;15 (v/v)). The ratio between the sample and the solvent used during extraction was 1:30 (w/v). Extraction was carried out in a waterbath shaker for 2 hours at 35 ° C, the sample was centrifuged for 20 minutes at 4 ° C, then the supernatant was separated for further analysis.

Sample extraction was carried out 0.5 ml was put into 2 test tubes, the first tube was filled with 4.5 ml of 0.025 M KCl buffer solution pH 1, while the second tube was filled with 4.5 ml of 0.4 M Na acetate buffer solution pH 4.5. The sample was incubated for 15 minutes in a dark room. Total anthocyanin measurement was carried out by measuring the absorbance of the sample with a UV-Vis spectrophotometer at a wavelength of 510 nm and 00 nm. The anthocyanin content was obtained from the difference in sample absorbance obtained at each solution pH and wavelength. The anthocyanin content was calculated as cyanide – 3 glucoside and calculated using the equation

$$\text{Antosianin} = \left(\frac{A \times MW \times DF \times 100}{\epsilon \times l \times W} \right) \times 100$$

h. Thickness

Edible film thickness was measured using a manual micrometer with an accuracy of 0.01 mm. The thickness value obtained was the average of measurements at 5 random points in mm (Warkoyo et al., 2014)

i. Solubility

Weigh the dried filter paper. The film sample was cut into 2x2 cm pieces, placed in 50 mL of distilled water, and soaked for 24 hours while stirring periodically. The solution was then filtered and the filter paper was dried at 50 °C for 3 hours. The amount of insoluble film was then weighed, (Silva et al., 2021). % Solubility can be calculated using the formula:

$$\% \text{ Kelarutan} = 100\% \left(\frac{w1 - w2}{w3} \right) \times 100\%$$

j. Transparency

The film was cut into 7×1 cm sizes and then inserted into a cuvette and placed in a spectrophotometer cell. The transmittance percentage (%) was measured using a UV-Vis spectrophotometer at a

wavelength of 600 Nm, (Zhao et al., 2022). The transparency of the edible film was calculated using the formula:

$$\text{Transparansi} = \frac{\log T}{K \text{etebalan}}$$

k. Water Vapor Transmission Rate /WVTR (Water Vapor Transmission Rate)

A test tube containing calcium chloride was covered with film. The tube was then weighed. The tube was placed in a desiccator saturated with saturated sodium chloride (RH 75%). The change in tube weight was then recorded and plotted as a function of time, (Piñeros-Hernandez et al., 2017). The WVTR calculation can be done using the following formula:

$$\text{WVTR} = \left(\frac{\text{slop}}{A} \right)$$

I. Compressive Strength

Compressive strength was measured using a Brookfield brand LFRA Texture Analyzer, (Santoso et al., 2018)

How the edible film compressive strength test works is:

1. The type of probe to be used for edible film is determined, namely the TA 7 60 mm type and a blade is used in testing the compressive strength of edible film.
2. The LFRA texture analyzer tool is set to:

Test : Cycle count

Trigger : 2g

Distance : 0.2 mm

Speed : 2 mm/s.

3. The probe is placed in place and the "start" button is pressed to begin pressing the edible film.
4. The edible film sample that has been cut to a size of 5×2 cm is placed under the probe and the probe will press the film until the magnitude of the probe force used appears on the screen.

Results And Discussion

a. Starch yield of purple uwi tubers (%)

The yield of modified starch was calculated as the percentage ratio between the weight of starch obtained after the modification process and the initial weight of starch before treatment. This calculation aims to determine how much starch yield increased due to the modification process. Meanwhile, the

yield of natural purple yam starch was obtained through the wet extraction method, namely by peeling, washing, crushing the tubers, then filtering and settling to obtain starch sediment which was then dried until it reached a stable condition. The analysis results showed that the yield of natural purple yam starch obtained through the wet extraction method was 20.42%. Meanwhile, the yield of purple yam starch modified with one treatment cycle increased significantly to 90.13%, and was even higher in the two modification cycles with a yield value of 98.16%. These data indicate a significant difference between natural starch and modified starch, where the modification treatment was able to increase the efficiency of starch recovery from purple yam tubers. The yield of yam tuber starch in this study showed a significant difference between natural starch and starch modified through the *autoclaving-cooling method*. While natural starch yielded only 20.42%, after modification, the yield increased significantly to 90.13% in one cycle and 98.16% in two cycles. These results indicate that the *autoclaving-cooling method* is effective in increasing the availability and release of starch granules from purple yam tuber tissue.

The results of this study align with those reported by (D. N. Faridah et al., 2022), who stated that *autoclaving-cooling treatment* can increase starch release and enlarge the starch fraction through molecular structural restructuring. The study also confirmed that the water ratio used during the autoclaving process contributes to the effectiveness of starch granule release. Similar results were also found for taro starch, where repeated autoclaving can break down the network structure, making the starch easier to extract (Wardana & Surono, 2022).

b. pH of Starch

The increase in pH value in starch modified with two autoclaving-cooling cycles indicates changes in the internal structure of starch granules. During the autoclaving process, starch granules undergo partial gelatinization due to heat and pressure treatment, followed by retrogradation during the cooling stage. This process can trigger the reorganization of amylose and amylopectin molecules, as well as the release of some functional groups that contribute to changes in the chemical properties of starch, including pH (Hoover, 2010). According to (Ashogbon & Akintayo, 2014), starch modification with the autoclaving-cooling method increases structural stability and reduces susceptibility to enzymatic degradation, so that the pH tends to increase compared to native starch.

Changes in pH in autoclaved-cooled modified starch are also closely related to the leaching process (the release of amylose molecules from the granules) and the formation of new hydrogen bonds during retrogradation. According to (Chung & Lai, 2023), repeated heating causes bond dislocation in starch granules and facilitates the release of more soluble amylose fractions, resulting in a relatively more neutral or slightly alkaline medium compared to native starch (S. Wang et al., 2020) stated that autoclaving-cooling treatment can increase the proportion of starch through the formation of stable amylose crystals. This process also changes the ionic characteristics of starch suspensions, which has implications for increasing pH values. These changes not only affect the chemical properties of starch but also determine its application in functional food products, especially those requiring stability at a certain pH range.

c. Water content (%)

This decrease in water content can be explained by the gelatinization and retrogradation processes. During the autoclaving stage, the crystalline structure of starch granules is disrupted by the influx of water molecules, while during the cooling stage, hydrogen bonds between amylose and amylopectin chains are reformed, resulting in a denser structure. This denser structure limits the granules' ability to absorb and store water, resulting in a decrease in water content after treatment (Hoover, 2001). The results of this study are consistent with those of (Deka & Sit, 2016), who reported that physical modification of starch through repeated heating and cooling treatments can reduce water content due to the formation of more stable intra- and intermolecular bonds. The *autoclaving-cooling cycle* increases the density of starch granules and increases the starch fraction, which results in a decrease in water-binding capacity. Therefore, the more treatment cycles, the lower the water content of the resulting starch.

d. Purple yam starch crude fiber

The increase in crude fiber content in the *autoclaving-cooling treatment* is closely related to the formation of the starch fraction. during the gelatinization and retrogradation processes. High-pressure heat treatment followed by cooling allows amylose molecules to form a crystalline structure that is difficult to hydrolyze, so this fraction resembles dietary fiber. These results are in line with the findings of (Sajilata et al., 2008), who stated that hydrothermal treatment can increase starch formation, which in proximate analysis is measured as a crude fiber fraction. (M. R. Faridah et al., 2023), who reported that an autoclaving-cooling cycle significantly increases the starch fraction, thereby increasing the dietary fiber value.

(Fuentes-Zaragoza et al., 2010), reported that repeated *autoclaving-cooling cycles* not only enhance amylose retrogradation but also promote the attachment of non-starch polysaccharides such as cellulose and hemicellulose to the starch matrix. This increases the crude fiber content, which can affect the functional quality of food ingredients. Higher crude fiber content has positive implications, including slowing the rate of starch digestion, lowering the glycemic index, and providing physiological benefits for gut health.

Table 1

Average results of soaking value, pH, water content, fiber content, of modified purple yam starch

Parameter	Treatment		
	Natural starch	1 cycle modified starch	2 cycle modified starch
yield (%)	20.42	90.13	98.16
pH	5.80 ^a	6.89 ^c	6.81 ^b
Water content (%)	8.72 ^b	7.51 ^a	7.26 ^a
Fiber Content (%)	5.22 ^a	6.24 ^b	6.56 ^c
Note : The average value is followed by a lowercase letter (subscript) that differs in direction. rows show significant difference (P < 0.05)			

e. Morphology of Starch Granules and Edible Fiber

Observations of the morphology of purple yam starch granules using a *Scanning Electron Microscope* (SEM) showed differences in granule structure before and after *autoclaving-cooling modification*. In natural starch granules (Figure a), the granule shape appeared relatively uniform with a round to oval morphology and a smooth surface and showed no structural damage. The granules appeared intact with clear edges, which is a characteristic of starch in its natural state. After one cycle of *autoclaving-cooling treatment* (Figure b), the granule morphology began to change, characterized by the appearance of small cracks, a rougher surface, and early indications of fragmentation. Some granules still maintained their oval shape, but their surfaces began to show irregularities. Meanwhile, after two cycles of autoclaving-cooling (Figure c), the granules experienced more intensive damage. Some granules appeared to break into small fragments, the surface structure became increasingly hollow and inhomogeneous, and there were indications of melting due to disruption. This phenomenon indicates that the higher the number of *autoclaving-cooling cycles*, the greater the degree of degradation of starch granule morphology that occurs.

Scanning Electron Microscope (SEM) images of purple yam starch, damage or rupture of the starch granule structure occurs due to physical treatment. In the autoclaving-cooling modification, damage occurs due to high-pressure heating cycles that trigger partial gelatinization, followed by cooling that causes retrogradation. The combination of these two processes causes the hydrogen bonds between amylose and amylopectin molecules to weaken, so that the internal structure of the granule becomes unstable and eventually experiences cracks, fragmentation, and even breaks into small particles. According to (Hoover, 2000), heat-cold treatment can trigger damage to starch granules by destroying crystalline regularity and expanding the amorphous region. The occurrence of damage is also associated with increased mobility of amylose molecules that exit the granule, resulting in granules with irregular surfaces. (da Rosa Zavareze & Dias, 2011) who stated that the higher the number of *autoclaving-cooling cycles*, the more intense the level of damage, characterized by broken granules, rough surfaces, and dominant amorphous structures.

f. Morphology of edible film granules

SEM observations of edible films based on purple yam starch show clear morphological differences between films made from natural starch, one *autoclaving-cooling cycle*, and two *autoclaving-cooling cycles*. In the edible film made from natural starch (a), the film surface appears inhomogeneous with the presence of cavities, small cracks, and incompletely dispersed granule agglomerations, indicating weak interactions between starch molecules in forming the film matrix. After one *autoclaving-cooling cycle* (b), the film surface appears smoother, cracks begin to decrease, and the granules appear to be more integrated into the matrix so that the film is more compact although there are still slight irregularities. Meanwhile, in the edible film with two *autoclaving-cooling cycles* (c), the film surface becomes denser, smoother, and more homogeneous, characterized by minimal cavities and cracks, indicating better integration of starch molecules and the formation of a stronger and more stable polymer network.

Scanning Electron Microscope (SEM) images, morphological observations of yam starch-based edible films show that autoclaving-cooling treatment has a significant effect on the level of granule damage and the homogeneity of the film surface. In edible films made from natural starch, the surface looks rough, inhomogeneous, and there are still agglomerations of granules that have not been fully dispersed. This condition indicates that natural starch granules that still maintain a semi-crystalline structure are difficult to integrate in the film matrix, resulting in a brittle film with many cavities. This phenomenon is in accordance with the explanation of (Tester et al., 2004), who stated that natural starch has strong hydrogen bonds in the crystalline region so that it is difficult to disrupt and disperse completely. After autoclaving-cooling, the film morphology becomes more compact and homogeneous. High-pressure heat treatment causes partial gelatinization, where amylose molecules exit the granules and act as polymer network-forming agents in the film matrix. The cooling process then triggers retrogradation, producing new, more stable hydrogen bonds and increasing the density of the film structure. (da Rosa Zavareze & Dias, 2011) reported that the *autoclaving-cooling method* was able to increase the proportion of starch through the gelatinization-retrogradation mechanism, which directly impacted changes in granule morphology.

autoclaving-cooling cycles increases, the surface of the edible film becomes denser, smoother, and free from cracks and cavities. This indicates more complete granule disruption and more optimal integration of starch molecules. (Ashogbon & Akintayo, 2014) explained that controlled disruption of starch granules can increase the homogeneity of the filmogenic solution, resulting in films with better mechanical and barrier properties. A similar study by (Thakur et al., 2019) also showed that edible films with a uniform surface and minimal pores have higher resistance to water vapor and gas permeability. Thus, *autoclaving-cooling modification* not only changes the morphology of starch granules but also improves the structural and functional qualities of the resulting edible film .

g. The color of the yam starch is purple

Table 2
Average results of purple cassava tuber starch color

Types of Starch	L*	a*	b*
Natural starch	67.49 ^b	5.95 ^a	13.20 ^a
1 cycle modified starch	66.84 ^b	5.96 ^a	13.14 ^a
2-cycle modified starch	65.59 ^a	6.39 ^b	14.35 ^b
Note : The average value is followed by a lowercase letter (subscript) that differs in direction. rows show significant difference (P < 0.05)			

Based on the research results, the average ANOVA analysis showed that the *autoclaving-cooling treatment* significantly affected the brightness value (*L*) of purple yam starch (F = 19.452; Sig. = 0.002). The highest *L value* was found in natural starch (67.49^b), followed by one-cycle modified starch (66.84^b), while the lowest value was found in two-cycle modified starch (65.59^a). The decrease in the *L value* in the two-cycle treatment indicates that the starch becomes darker due to the degradation of anthocyanin pigments and non-enzymatic browning reactions during the pressurized heat treatment. Surawan et al. (2024) reported that the *autoclaving-cooling cycle* reduces the brightness of starch due to pigment damage and changes in granule structure that affect light reflection. This finding is consistent with the results of the study, where the more treatment cycles, the lower the *L value*.

On the *a parameter*, ANOVA showed a highly significant effect (F = 425.049; Sig. = 0.000). The highest *a value* was shown by the two-cycle modified starch (6.39^b), while the natural starch (5.95^a) and one-cycle modified starch (5.96^a) had lower *a values*. This indicates that the two-cycle treatment increased the intensity of the red color in purple yam starch. (M. R. Faridah et al., 2023) stated that *autoclaving-cooling treatment* accelerated the degradation of anthocyanin pigments, which caused a color shift towards brownish red. This condition strengthens the finding that increasing the number of autoclaving-cooling cycles enlarges the color change, marked by a significant increase in the *a value*.

The ANOVA results for the *b parameter* also showed a very significant effect (F = 251.211; Sig. = 0.000). The highest *b value* was obtained in two-cycle modified starch (14.35^b), while the lowest values were found in one-cycle modified starch (13.14^a) and natural starch (13.20^a). The increase in the *b value* indicates a color shift towards yellow due to the degradation of anthocyanin pigments and mild Maillard reactions that occur during thermal treatment. (Surawan et al., 2024) also reported a similar phenomenon in sorghum starch, where autoclaving-cooling treatment increased the *b value* as a result of changes in the structure of pigments and non-starch components. Thus, the more modification cycles, the more the starch color shifts towards yellow.

h. The edible color of the purple yam tuber flesh

Table 3
Average results of edible color of purple yam tubers

Treatment (Interaction of t and l)	L*	a*	b*
t0l1	73.05 ^b	5.17 ^d	17.96 ^d
t0l2	71.96 ^a	4.85 ^{CDs}	18.47 ^e
t0l3	72.07 ^a	4.93 ^d	18.63 ^e
t1l1	73.91 ^{bc}	3.56 ^a	17.33 ^c
t1l2	77.47 ^d	4.34 ^{bc}	18.91 ^e
t1l3	72.62 ^{ab}	4.42 ^{bc}	14.78 ^a
t2l1	74.05 ^{bc}	4.47 ^{bc}	17.54 ^{cd}
t2l2	74.80 ^{cd}	4.37 ^{bc}	18.59 ^e
t2l3	71.99 ^a	4.06 ^{ab}	16.35 ^b
Note : The average value is followed by a lowercase letter (subscript) that differs in direction. rows show significant difference (P < 0.05)			

Changes in the L value of modified purple yam starch edible film indicate that *autoclaving-cooling treatment* affects the film's brightness level. Increased L values in several treatments indicate brighter films due to gelatinization and retrogradation, which produce a more homogeneous starch matrix, allowing light to be more easily reflected. (M. R. Faridah et al., 2023) reported that autoclaving-cooling can change the crystalline structure and functional properties of starch, including increasing product stability and transparency (Frontiers in Nutrition,). These results are also in line with (Ke et al., 2024) who found that thermal treatment of purple yam-based films causes an increase in lightness due to the partial degradation of anthocyanin pigments. Thus, variations in L values in this study can be attributed to a combination of starch restructuring and natural pigment degradation.

The a value is more influenced by the type of modified starch than by pigment concentration, indicating that red stability is highly dependent on matrix conditions. *Autoclaving-cooling* causes amylose chain reorganization, allowing new hydrogen bonds to form that protect anthocyanins, but heat treatment also has the potential to accelerate pigment degradation. (Chen et al., 2019) reported that heating can reduce the stability of purple sweet potato anthocyanins, indicated by a decrease in red color intensity. Furthermore, microstructures in starch can also trigger anthocyanin color shifts from red to purple or bluish. This reinforces the finding that the modified starch matrix is more important for red color expression than simply increasing pigment concentration.

The b value is significantly influenced by starch treatment, pigment concentration, and their interaction, indicating that the yellow-blue color shift is the result of a combination of these factors. The shift toward

yellow is often associated with the degradation of anthocyanins to brown-yellow products due to heat treatment. (Sohany et al., 2021) reported that starch-based films with added purple sweet potato anthocyanins experienced an increase in b value as pigment degradation occurred during storage. Meanwhile, (da Rosa Zavareze & Dias, 2011) stated that heat-moisture treatments, including autoclaving, can modify the molecular structure of starch and have direct implications for edible appearance. Thus, the high significance of the treatment interaction on the b value indicates the important role of starch condition in maintaining or shifting the film color toward yellow.

i. Anthocyanin starch and edible flem of purple yam tuber

Table 4
Average yield of anthocyanin in starch and edible pulp of purple yam tubers

Types of Starch	Starch anthocyanin	Edible anthocyanin flem
Natural starch	3.17 ^a	8.07 ^a
1 cycle modified starch	5.82 ^b	11.12 ^b
2-cycle modified starch	5.86 ^b	11.10 ^b
Note : The average value is followed by a lowercase letter (subscript) that differs in direction. rows show significant difference (P < 0.05)		

The increase in anthocyanin content in modified starch can be explained by the phenomena of gelatinization and retrogradation. The autoclaving process causes the starch granules to rupture and the amylose-amylopectin structure to disrupt, allowing the anthocyanin pigments previously trapped within the granules to be more easily released. After cooling, retrogradation occurs, resulting in a new, more open starch network. This process increases the availability of pigments for extraction. These findings suggest that *autoclaving-cooling treatment* can increase the bioavailability of phenolic compounds in food (Zhang et al., 2015). Furthermore, research on purple yam by (Deka & Sit, 2016) also showed that although anthocyanins are sensitive to heat, under certain conditions thermal treatment actually triggers the release of pigments from the network, thereby increasing the measured content. Thus, autoclaving-cooling not only functions to modify the starch structure but also contributes to increasing the functional value of the material.

The increase in anthocyanin content in modified starch-based edible films can be explained by the ability of the retrograded starch structure to form a denser and more homogeneous network. This structure allows anthocyanin pigments to be dispersed more evenly and protected from oxidative degradation. According to (Dai et al., 2019) modified starch can interact with phenolic compounds, thereby improving bioactive retention in edible films. Furthermore, the presence of anthocyanins in edible films not only increases their functional value as antioxidants but also provides attractive natural colorants. Huang et al. (2019) emphasized that edible films with added natural pigments can improve the oxidative stability of packaged food products. Therefore .

Table 5

Average results of the values, thickness, transparency, solubility, wvtr, compressive strength, and crude fiber content of edible films from modified purple yam starch .

Treatment	Observation Parameters						
	Thickness	Transparency	Solubility	Wvtr	Compressive strength	pH	Crude fiber
t0l1	0.21 ^b	6.23 ^{ef}	67.22 ^a	11.08 ^c	207.00 ^a	5.72 ^a	3.95 ^a
t0l2	0.23 ^c	6.02 ^{de}	78.20 ^c	7.39 ^b	217.67 ^{ab}	5.72 ^a	3.86 ^a
t0l3	0.24 ^d	5.61 ^{bc}	67.28 ^a	3.69 ^a	239.00 ^c	5.72 ^a	4.65 ^c
t1l1	0.21 ^b	6.32 ^f	78.63 ^c	7.39 ^b	218.33 ^b	6.00 ^b	4.18 ^{ab}
t1l2	0.22 ^c	5.78 ^{cd}	78.60 ^c	7.39 ^b	236.33 ^c	6.12 ^{CDs}	4.65 ^c
t1l3	0.24 ^d	5.45 ^{ab}	78.43 ^c	3.69 ^a	281.33 ^a	6.14 ^d	4.70 ^c
t2l1	0.20 ^{ab}	6.65 ^f	70.81 ^b	7.39 ^b	221.67 ^d	6.10 ^c	4.49 ^{bc}
t2l2	0.23 ^c	5.64 ^{bc}	70.80 ^b	7.49 ^b	251.67 ^d	6.11 ^{cd}	4.35 ^{bc}
t2l3	0.24 ^d	5.27 ^a	71.02 ^b	3.69 ^a	292.33 ^f	6.17 ^e	4.48 ^{bc}
Note : The average value is followed by a lowercase letter (subscript) that differs in direction. rows show significant difference (P < 0.05)							

j. Edible film thickness

Edible film thickness is an important parameter because it is related to the film's mechanical properties and protective function. The results of this study indicate that increasing the concentration of starch solution significantly affects film thickness. The higher the concentration of added solids, the greater the viscosity of the solution, resulting in a thicker film (Praseptiangga et al., 2022). Furthermore, *autoclaving and cooling* purple yam starch results in structural changes through gelatinization and retrogradation, which increase matrix density and promote film formation with more uniform thickness (Zhu et al., 2021).

The interaction between modified starch and solution concentration was also significant, indicating that starch modification can enhance the effect of concentration on thickness. This study aligns with (Zhang

et al., 2008) that heat-hydrothermal treatment of starch can enhance film-forming ability due to the reorganization of amylose and amylopectin molecules. Thus, these results demonstrate that edible films from modified purple yam starch are influenced not only by the formulation composition but also by the starch treatment conditions. The thickness produced in this study (0.19–0.24 mm) is comparable to other starch-based edible films, such as corn and cassava starch, which are reported to range from 0.17–0.25 mm (Ramadoss et al., 2025). This indicates that purple yam starch modified through autoclaving–cooling has the potential to be a raw material for edible films with competitive physical qualities compared to other commercial starch sources.

k. Edible film transparency

This study shows that the transparency properties of edible films are not solely influenced by a single factor, such as starch type or solution concentration, but rather by the simultaneous interaction of both. In some treatment combinations, increasing the solution concentration is able to form a denser and more homogeneous film matrix, resulting in increased light transmission and higher film transparency. Conversely, certain combinations actually trigger retrogradation and the formation of crystalline structures in modified starch, which act as light barriers and thus reduce transparency. These results are in line with the findings of Singh et al. (2022) who reported that polymer concentration and modification conditions are important factors determining the optical properties of starch-based films.

Physical modification through autoclaving–cooling has been shown to alter the molecular structure of starch through repeated gelatinization and retrogradation processes. This treatment increases crystallinity, reduces granule size, and modifies hydrogen bonding patterns, ultimately affecting the optical properties of the film. (Paixão e Silva et al., 2021) confirmed that this method is effective in improving the functional properties of starch, including its application in edible film production. In this study, the combination of treatments 7, 4, 1, and 2 produced the highest transparency value, reflecting the formation of a more homogeneous film matrix due to optimal interactions between the modified starch and the solution concentration. In addition, the natural anthocyanin pigment from purple yam tubers also plays a role in determining the transparency value of the film. This pigment absorbs light and can therefore reduce optical transmission, but its effect is highly dependent on the level of dispersion in the film matrix. (Yue et al., 2024) reported that purple yam-based films maintained good transparency when the pigments were homogeneously dispersed, whereas at high pigment concentrations, aggregation tended to occur, leading to increased light scattering. Thus, the results of this study confirm that the optimization of the transparency of edible films based on purple yam starch is related to the starch modification, solution concentration, and distribution of natural pigments.

l. Edible Film Solubility

The solubility of edible films in various treatment combinations shows that the interaction between the type of modified purple yam starch and the solution concentration significantly determines the film's functional properties. The *autoclaving-cooling process* causes damage to starch granules, followed by gelatinization and partial retrogradation, which changes the starch structure, especially in the

distribution of amylose and amylopectin. These changes have direct implications for the formation of the film matrix, where at certain concentrations the resulting structure is looser and more porous, thus facilitating the penetration of water molecules, ultimately increasing solubility. Conversely, other treatment combinations actually strengthen the interactions between amylose and amylopectin chains, resulting in a denser matrix structure that is more resistant to dissolution.

The high solubility in some combinations indicates that the autoclaving-cooling process is capable of releasing the soluble amylose fraction, which contributes to the improvement of the hydrophilic properties of edible films. This is in line with the findings of (Dai et al., 2019) who emphasized that the proportion of free amylose plays a significant role in regulating the water permeability and solubility of starch-based films. Furthermore, (X. Wang et al., 2024) also reported that the autoclaving-cooling treatment accelerates the reorganization of starch molecules, making hydroxyl groups more available to interact with water molecules, ultimately affecting solubility. In addition to structural factors, the solution concentration level also plays a significant role. Increasing the concentration can produce thicker films due to the increased amount of matrix-forming solids. This condition is consistent with the results of the study by (Gómez-Contreras et al., 2021) who explained that increasing solids in the film solution limits water diffusion, thereby reducing solubility.

m. Water Vapor Transmission Rate (WVTR)

The results showed that the water vapor transmission rate (WVTR) of purple yam starch-based edible films was significantly influenced by the type of modified starch, the solution concentration, and the interaction between the two. Autoclave-cooling treatment caused changes in the starch structure through gelatinization and retrogradation processes, resulting in a denser and more stable polymer matrix. This more compact structure was able to reduce the number of micropores in the film, thus limiting the water vapor diffusion path. This condition was seen in certain treatment combinations (3, 6, and 9) which produced the lowest WVTR (3.69 g/m²·day), indicating an increase in water vapor barrier properties. This finding is in line with the report of (Fatima et al., 2024) which stated that starch modification through pressure heat treatment can increase crystallinity and intermolecular hydrogen bonding, thereby reducing the moisture permeability of starch-based films.

Furthermore, solution concentration plays a crucial role in determining the barrier properties of the film. At low concentrations, the resulting film tends to be thin and inhomogeneous, facilitating the diffusion of water molecules. This is reflected in the treatment with the highest WVTR (11.08 g/m²·day), indicating that the polymer content is insufficient to form a cohesive network. Conversely, increasing the concentration to the optimum point produces a film with a denser and more uniform structure, resulting in a decrease in WVTR. This phenomenon is consistent with research by (Nogueira et al., 2019), who reported that increasing starch concentration in the film solution limits porosity and increases matrix density, thereby decreasing water vapor permeability. However, excessively high concentrations can excessively increase solution viscosity and cause film thickness non-uniformity, which in some conditions can actually affect the water vapor diffusion path.

The interaction between the type of modified starch and the solution concentration shows that the effectiveness of purple yam starch modification in controlling WVTR is not a single factor, but rather is influenced by a combination of both. Each type of modification produces different characteristics in terms of molecular distribution and degree of recrystallization, so that only at certain concentrations does the resulting film structure become dense enough to optimally retain water vapor. This finding is consistent with the results of research by (Parera & Gusriani, 2021), which showed that edible films based on native purple yam starch have a relatively high WVTR, but starch modification can significantly reduce it by forming a more ordered internal structure. Recent studies also emphasize that the main weakness of starch-based films is their hydrophilicity, making physical modification such as autoclaving–cooling an effective strategy to improve moisture barrier properties (Kupervaser et al., 2023).

o. Compressive Strength

The significant interaction between modified starch and starch concentration indicates that the *autoclaving-cooling process* plays a crucial role in shaping the mechanical characteristics of edible films. This process causes granules, gelatinization, and the re-formation of the crystalline structure (retrogradation), resulting in stronger molecular bonding in purple yam starch when combined with starch concentration. The higher the concentration, the tighter the hydrogen bonds between the polymer chains, thereby increasing the film's resistance to compressive forces. Furthermore, the presence of bioactive compounds in purple yam, such as anthocyanins, can also influence intermolecular interactions. Anthocyanins play a role in strengthening the film network through the formation of non-covalent bonds with starch molecules. This is in line with the findings of Abdillah et al. (2021) who reported that natural pigments can contribute to increasing the mechanical strength of starch films through polymer-pigment interactions.

The results of this study are consistent with the report of (Ramos et al., 2016) which showed that *autoclaving-cooling* cycles increased starch crystallinity and improved the mechanical properties of biopolymer-based films. They also stated that the combination of starch modification and solution concentration produced a denser and more homogeneous polymer matrix, thereby strengthening the mechanical properties of edible films. Similar findings were presented by (Shanbhag et al., 2023), who found that the interaction between starch modification and solution concentration was able to improve polymer distribution and mechanical resistance of films. Thus, the results of this study confirm that the combination of *autoclaving-cooling modified purple yam starch* with the appropriate concentration is able to produce edible films with optimal compressive strength.

p. Edible crude fiber from purple cassava tuber flesh

It can be seen that the average fiber content of edible films increases with the combination of *autoclaving-cooling* and varying wax concentrations. For example, in the unwaxed control (t0), the average fiber content was around 3.95 g in the first replication, while in the combination of t1 and I3 with

concentration values, the fiber content increased to around 4.70 g. This indicates an interaction effect, not only *autoclaving-cooling*, and not only starch concentration, but also the combination of both that increases the fiber content of edible films. In general, these results confirm that the better the interaction between modified starch and starch concentration, the higher the crude fiber content of the resulting edible film. This is important because the optimal crude fiber content not only affects the nutritional quality, but also the shelf life and mechanical stability of edible films (Luchese et al., 2017).

According to (Septia & Supriadi, n.d.), hydrothermal treatments such as *autoclaving-cooling* can increase the formation of starch, which acts as dietary fiber. Meanwhile, research by (Carpiné et al., 2015) shows that the addition of hydrophobic components such as starch to edible films can improve this by filling the empty spaces between polymer chains. The combination of these two mechanisms explains why the interaction between starch type and starch concentration produces significant differences in the crude fiber value of edible films. Thus, the right combination of treatments has the potential to produce functional edible films with higher dietary fiber content and better mechanical characteristics.

q. pH Eibel Flem

The pH value of the edible film, which is in the neutral range (5.7–6.2), indicates that the modification of purple yam starch through the *autoclaving-cooling method* is able to produce a polymer matrix that is relatively stable against ionization changes. The autoclaving process at high temperature and pressure followed by repeated cooling triggers starch retrogradation and the formation of a partial crystalline structure that can affect the film's buffering capacity. These changes have implications for the film's ability to retain hydrogen ions, so the final pH tends to increase compared to unmodified starch. These results are consistent with the findings of (Paixão e Silva et al., 2021) who reported that *autoclaving treatment* can modify the functional properties of tuber starch, including its chemical properties, through the reorganization of amylose and amylopectin molecules.

Furthermore, the presence of anthocyanin pigments in purple yam also plays a role in the pH stability of the film. Anthocyanins are pH-sensitive and can undergo structural changes depending on ionization conditions. However, the interaction between anthocyanins and the modified starch matrix allows for the formation of hydrogen bonds and van der Waals forces that stabilize the pigments and reduce pH fluctuations. This is in line with the report by (Ke et al., 2024), which showed that starch-based films with natural pigment extracts were able to maintain pH stability while showing potential as indicators of changes in food quality. Thus, edible films based on modified purple yam starch not only offer good chemical stability but also have the potential to be applied as intelligent packaging to detect changes in food product quality.

Furthermore, the pH stability of the films produced in this study is comparable to the results of (Rahmadhia et al., 2023) who used tapioca-based films with the addition of purple sweet potato anthocyanin extract. They reported that the resulting indicator film was able to show a clear pH response while maintaining stability in the near-neutral pH range. This indicates that modified purple sweet potato

starch has a dual function, namely as a polymer matrix with good mechanical and barrier properties, as well as a pH buffer medium suitable for food applications.

Conclusion

This study proves that the modification of purple yam starch (*Dioscorea alata* L.) through repeated autoclaving-cooling methods with varying starch concentrations significantly affects the physical, chemical, and functional characteristics of the starch and the quality of the resulting edible film. Autoclaving-cooling treatment can increase starch yield to nearly 98%, reduce water content, increase pH, and enrich crude fiber and anthocyanin content. The results of morphological observations show that the more autoclaving-cooling cycles, the more intensive the damage to starch granules that occurs, which has implications for the formation of a more homogeneous, dense, and stable film structure. At the edible film level, the interaction between the type of modified starch and the solution concentration produces a significant effect on thickness, transparency, solubility, color, fiber content, as well as mechanical properties and water vapor barrier properties. The right combination of treatments is proven to produce edible films with better functional properties, characterized by low WVTR, good transparency, and high compressive strength. Thus, purple yam starch modified through autoclaving-cooling has the potential to be developed as an alternative raw material in the production of functional edible films, which not only has competitive physical and mechanical characteristics, but also added value from the content of natural bioactive pigments.

Declarations

1. Funding Declaration in the manuscript

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2. Data Availability

All data generated or analyzed during this study are included in this published article

3. Competing Interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Author Contribution

Author 1 (the lead author) contributed to the research conceptualization, methodological design, supervision, experimental implementation, data collection, and data analysis, as well as writing the initial manuscript. Author 2 contributed to data validation and interpretation. Author 3 reviewed and edited the manuscript. Author 4 assisted with the literature review and revision of the manuscript. All authors have read and approved the final manuscript for publication.

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Figures

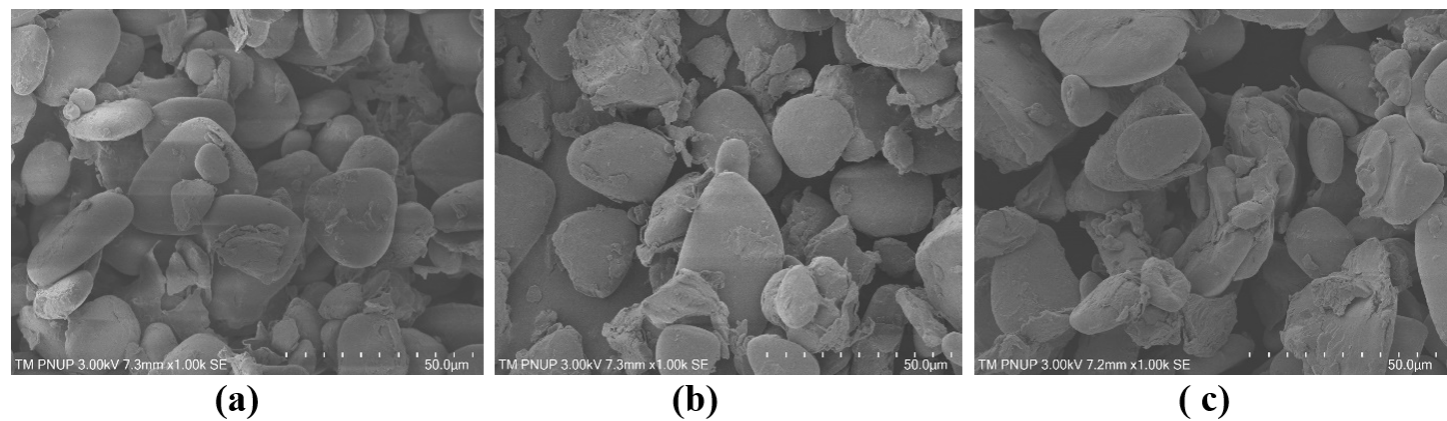


Figure 1

Granule shape of SEM test results at 1000× magnification for (a) purple yam starch without modification; (b) purple yam starch modified with 1 cycle, (c) purple yam starch modified with 2 cycles

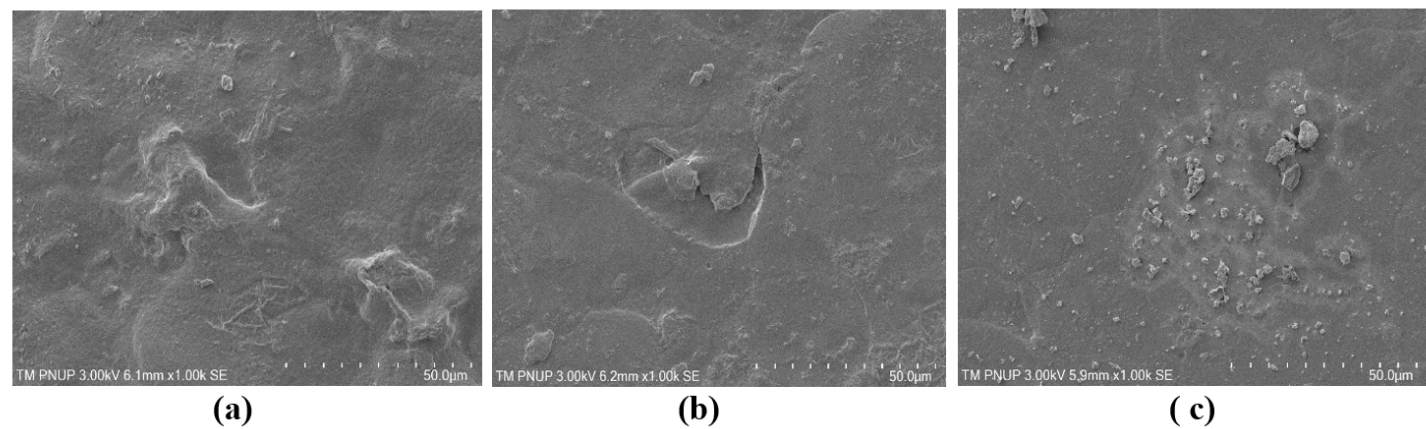


Figure 2

Granule shape of SEM test results at 1000× magnification for (a) edible film without modification; (b) edible film with 1 cycle modification ; (c) edible film with 2 cycle modification