

## Characteristics of *Dioscorea esculenta* Starch Exposed to Citric Acid and Autoclaving-Cooling Treatment

Andriana Puspitasari<sup>1,2</sup>, Yudi Pranoto<sup>1,✉</sup>, Priyanto Triwitono<sup>1</sup>, Dwi Larasatie Nur Fibri<sup>1</sup>

<sup>1</sup> Department of Food and Agricultural Product Technology, Faculty of Agricultural Technology, Universitas Gadjah Mada, Yogyakarta, INDONESIA

<sup>2</sup> Department of Fisheries and Marine Resources Management, Faculty of Fisheries and Marine Science, Universitas Brawijaya, Malang, INDONESIA

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Corresponding Author:

✉ [pranoto@ugm.ac.id](mailto:pranoto@ugm.ac.id)  
(Yudi Pranoto)

### ABSTRACT

*Starch is an abundant carbohydrate reserve found in plants, such as Dioscorea esculenta. Starch can be a staple food and energy source. However, native starch has limitations, required modification processes. The research investigated effect of citric acid and autoclaving-cooling in several cycles for its starch characterization. Starch was extracted and pretreated with citric acid (0.1 M) for 6 h at a ratio of starch and water of 1:4 (w/v). Starch was then modified by autoclaving-cooling using different cycles (1, 2, 3, 4, and 5). Analyses included recovery yield, proximate composition, amylose content, water or oil holding capacity, solubility, swelling power, and color. The results showed no significant difference in the recovery yield, decreased protein, and increased amylose. Autoclaving-cooling modification in Dioscorea esculenta starch enhances the water holding capacity (WHC), oil holding capacity (OHC), and solubility, while reducing its swelling power. In terms of the color of starch after modification by autoclaving-cooling, there was a decrease in lightness (L\*) and the whiteness index (WI) values. The results suggested that starch produced with two cycles of autoclaving-cooling was better, as indicated by an increase in amylose content of 25.14%. These findings are expected to provide alternative sources of functional foods and contribute to the utilization of local food ingredients.*

## 1. INTRODUCTION

*Dioscorea* represents one of the most extensive genera within the family. *Dioscorea*, commonly known as true yams, includes *Dioscorea alata* and *Dioscorea esculenta*, two widely cultivated species. *Dioscorea esculenta* has cylindrical, spiny, and tangled stems (Jayakody *et al.*, 2007) and is characterized by its oval and small shape, length of 5-10 cm, and 2.7-4 cm in diameter, weight 50 to 150 g, light brown skin, exhibits small root development, and contains clean white flesh. *Dioscorea esculenta* has an inulin content of 14.77% (db) in fresh yam, with a moisture content of 84.57%, and an ash content of 1.11% (Winarti *et al.*, 2011). *Dioscorea esculenta* tuber has been examined for its flour's functional and structural properties (Varshney *et al.*, 2024), starch's physicochemical properties (Jayakody *et al.*, 2007), nutritional components from different cultivation locations (Sato *et al.*, 2024), and changes in nutritional and antinutritional components caused by cooking (Wanasundera & Ravindran, 1992). Nonetheless, few studies has been conducted to explore the effect of starch modification, such as citric acid treatment and different autoclaving-cooling cycles on *Dioscorea esculenta*.

Starch contain amylopectin and amylose, are glucans that vary in structural and molecular weight. Amylose displays a low molecular weight, but amylopectin exhibits a higher molecular weight (Cornejo-Ramírez *et al.*, 2018). The amounts of amylose and amylopectin in starch range from 25-30% and 70-75%, respectively (Zhao *et al.*, 2018). The starch properties are dependent on its plant origin, and modification is expected to improve its qualities and

increase its usefulness. Modifications applied to cassava and potato starch by organic acids and heat-moisture treatments caused changes in physical properties and increased resistant starch levels (Pham *et al.*, 2017), while modifications to ginseng starch with citric acid-autoclaving resulted in differences in physicochemical characteristics and in vitro digestibility (Cui *et al.*, 2024).

Starch hydrolysis with acid induces randomized cleavage of  $\alpha$ -1,4-glycosidic or  $\alpha$ -1,6-glycosidic bonds, resulting in shorter amylose chains. Physical treatments, such as autoclaving-cooling, can improve the stability of starch granules and facilitating digestive enzyme activity (Nurhayati *et al.*, 2022). Autoclaving-cooling is one of starch modification methods consisting of high-pressure heating followed by a cooling stage (Adilla *et al.*, 2020). This process occurs at a temperature of around 100 °C and is accompanied by pressure, with cooling temperatures ranging from 4 to 5 °C. A amount of water is required for this process to form amylose chains and help them connect, thus creating stable hydrogen bonds (Wijanarka *et al.*, 2020). The modification process carried out forms retrograded short chain starch and will bind to each other to form a double helix structure (Frista Silitonga *et al.*, 2021). Gelatinization breaks down the structure of amylose and amylopectin within the starch complex during the process. The gelatinization process enhances the availability of starch in digestive enzymes. The use of cooling treatment will facilitate the development of type 3 resistant starch. Recrystallization forms hydrogen bonds between type 3 resistant starch molecules (Ogbo & Okafor, 2015).

Starch modification using autoclaving and cooling showed several changes, such as amylose content. Arenga starch modified with one and two cycles of autoclaving and cooling showed increased amylose content compared to native starch. The amylose content showed the same value as native starch when three cycles were used (Ratnaningsih & Arovah, 2025). Cowpea starch modified by autoclaving-cooling showed the highest amylose content with one cycle (Ratnaningsih *et al.*, 2020). Based on the results of current research, the impact of the autoclaving-cooling cycle on starch that has previously been treated with citric acid hydrolysis has not been discussed. The aim of the research was to investigate how citric acid and autoclaving-cooling treatment affected the properties of *Dioscorea esculenta* starch, such as recovery yield, proximate composition, amylose content, water or oil holding capacity, solubility, swelling power, and color. The findings can provide thorough information regarding the properties of *Dioscorea esculenta* starch after it has been treated (citric acid and autoclaving-cooling) and affect the autoclaving-cooling cycle on starch, besides that, it also provides information regarding future development and use in the food industry.

## 2. MATERIALS AND METHODS

### 2.1. Material and Tools

*Dioscorea esculenta* was collected August to October 2022 from a local farmer in Kulon Progo, Special Region of Yogyakarta, Indonesia. All chemical reagents used in this research were of analytical grade. The tools for sample preparation and analysis are blender (Miyako, BL-152 GF/PF-AP, Indonesia), autoclave (Zealway GR 110DR, Zealway Instrument Inc, China), cabinet dryer, sieve (EML 200 Pure, Haver and Boecker OHG, Germany), analytical balance (Ohaus PR224, Ohaus Instrument, USA), oven (Mettler, Germany), hotplate magnetic stirrer (IKA C-MAG HS 7, IKA, Germany), pH meter (Mettler-Toledo, Mettler-Toledo GmbH Process, Switzerland), spectrophotometer UV-Vis (Genesys 10S UV-VIS, Thermo Fisher Scientific corporation, USA), vortex (Vortex Maxi Mix II, Thermo Fisher Scientific corporation, USA), centrifuge (SorvallTM ST 8 Centrifuge, Thermo Fisher Scientific corporation, USA), waterbath (Mettler Water bath WNB 29, Germany), and chromameter (CR-400, Konica Minolta, Japan).

### 2.2. Sample Preparation

*Dioscorea esculenta* starch was extracted following Mutmainah *et al.* (2021) with some modification. That resulting starch was identified as native starch (NS). Before autoclaving and cooling, NS was treated with 0.1 M citric acid at a 1:4 (w/v) ratio for 6 h, followed by drying and sieving to 80 mesh. The selection of methods for pretreatment was determined by Puspitasari *et al.* (2025), who determined the optimal concentration and time of hydrolysis.

After pretreatment, starch modifications were carried out using an autoclaving-cooling method, based on Ratnaningsih *et al.* (2020) with modifications. Distilled water (60 mL) was added into starch (20 g) (1:3, w/v) and was homogenized. The homogenized starch was subjected to autoclaving at 121 °C for 20 min and then cooled at room

temperature. The following step required cooling the mixture at 4 °C for 24 h. The autoclaving-cooling process was referred to as cycle 1. The autoclaving-cooling procedure was conducted for 2, 3, 4, and 5 cycles. The following is a classification of the starches from the different treatments: S1: one autoclaving-cooling cycle; S2: two autoclaving-cooling cycles; S3: three autoclaving-cooling cycles; S4: four autoclaving-cooling cycles; S5: five autoclaving-cooling cycles. The drying was carried out at 50 °C for 12-24 h, followed by grinding and sieving at 80 mesh.

### 2.3. Analysis

#### 2.3.1. Recovery yield of starch

The recovery yield value of starch was calculated using Equation (1):

$$\text{Recovery yield(\%)} = \frac{\text{Starch weight after autoclaving - cooling}}{\text{Starch weight before autoclaving - cooling}} \times 100 \quad (1)$$

#### 2.3.2. Proximate composition

The proximate composition is an essential analysis for determining both native and starch modified by autoclaving-cooling. Proximate analysis was performed following the methodology described by the Association of Official Analytical Chemists (AOAC) procedure, which included moisture, protein, lipid, and ash content, with carbohydrate content calculated by difference.

#### 2.3.3. Amylose content

Samples of 100 mg were mixed in 1 mL of 95% ethanol and 9 mL of 1 N NaOH. The mixture was heated for 10 min until gel formation occurred. The resulting gel was transferred to a volumetric flask and diluted to a final volume of 100 mL. Subsequently, 5 mL of starch solution was mixed with 1 mL of 1 N acetic acid and 2 mL of iodine reagent. This mixture was then diluted with distilled water to a total volume of 100 mL. The solution required incubation for 20 min before the analysis at a wavelength of 625 nm (Sonia *et al.*, 2019).

#### 2.3.4. Water or oil holding capacity

The water holding capacity (WHC) and oil holding capacity (OHC) of the samples (2 g) were mixed with 10 mL of distilled water or sunflower oil, as indicated by Raza *et al.* (2021). The sample was homogenized with water or oil using vortex for 5 min and then centrifuged at 3000 x g for 10 min. Following centrifugation, the supernatant was removed, and the wet pellet was analyzed. The WHC or OHC values were calculated with Equation (2):

$$\text{WHC or OHC} = \frac{\text{Wet pellet weight} - \text{Initial dry weight}}{\text{Initial dry weight}} \quad (2)$$

#### 2.3.5. Solubility and swelling power

Solubility and swelling power were determined using the method provided by Zavareze *et al.* (2010), with slight modifications. The samples (0.2 g) were mixed with 10 mL of distilled water. The suspensions were heated for 30 min at 60, 70, and 80 °C. The suspensions were manually homogenized, cooled to room temperature, then centrifuged at 1000 x g for 20 min. The supernatants were then transferred to a weighing bottle and kept at 105–110 °C until a constant weight was achieved. The solubility and swelling power values were derived using Equation (3) and (4):

$$\text{Solubility (\%)} = \frac{\text{Weight of dry supernatant}}{\text{Initial weight}} \times 100 \quad (3)$$

$$\text{Swelling power (g/g)} = \frac{\text{Weight of sediment}}{\text{Initial weight}} \quad (4)$$

#### 2.3.6. Color

The starch color was measured using a colorimeter (CR-400, Konica, Japan) as described by Pejcz *et al.* (2023). The CIE lab values (L, a\*, and b\*) were used to determine the starch color, represented as the total color difference ( $\Delta E$ )

from native starch, chroma, and hue. The whiteness index (WI) was calculated by using formula in [Ratnaningsih et al. \(2020\)](#). The  $\Delta E$ , WI, chroma, and hue value was determined using Equation (5), (6), (7), and (8):

$$\text{Whiteness index} = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \tag{5}$$

$$\text{Chroma (C)} = \sqrt{a^{*2} + b^{*2}} \tag{6}$$

$$\text{hue (h}^0\text{)} = \arctan\left(\frac{b}{a}\right) \tag{7}$$

$$\Delta E = \sqrt{(L^* - L_o^*)^2 + (a^* - a_o^*)^2 + (b^* - b_o^*)^2} \tag{8}$$

## 2.4. Statistical Analysis

The data are shown as the mean  $\pm$  standard deviation (SD). The statistical analysis utilized a one-way analysis of variance. The differences between means were analyzed using Duncan’s Multiple Range Test (DMRT) at  $\alpha = 0.05$ . The data were then analyzed using SPSS (version 25.0, SPSS Inc., Chicago, IL, USA).

## 3. RESULTS AND DISCUSSION

### 3.1. Recovery Yield of Starch

Figure 1 exhibits the recovery yield for native starch and starch modified by citric acid and autoclaving-cooling. Calculations of recovery yield were performed to compare the starch used and the starch produced. The starch recovery yield during the autoclaving-cooling with different cycles ranged from 95.55-98.01%. The results indicate no significant difference between native starch and starch modified using autoclaving-cooling, with differing cycles producing similar results. [Surendra Babu et al. \(2016\)](#) demonstrated that the recovery yield of sweet potato starches modified with citric acid was 90.73%. [Dutta et al. \(2011\)](#) found that starch produced from jackfruit seeds had a recovery yield between 73.22% and 86.32%. [Rahayu et al. \(2020\)](#) showed that breadfruit starch, when autoclaved and cooled for three cycles, had a recovery yield between 73.93% and 97.22%. The results indicate that the autoclaving-cooling procedure for modifying starch do not significantly reduce the sample used.

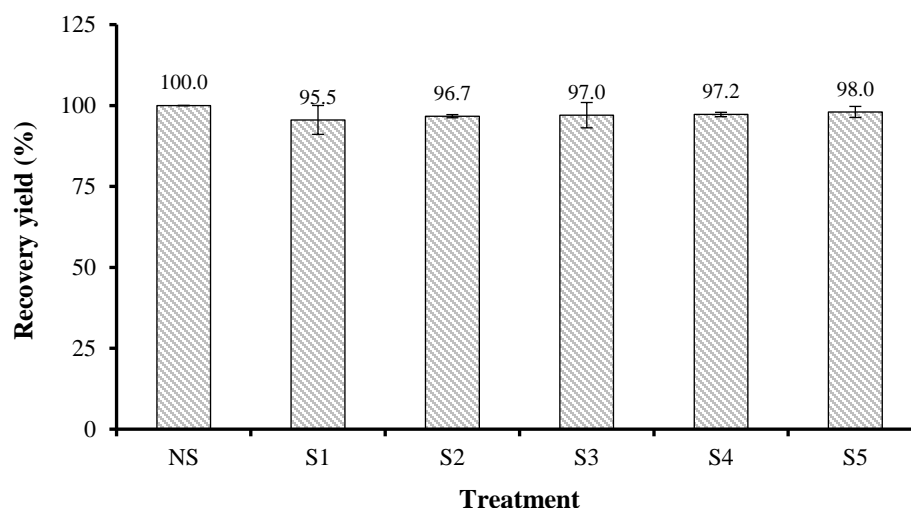


Figure 1. Recovery yield of starch. [Note: All values are presented as mean  $\pm$  SD. Same superscript letter indicate no significant differences based on DMRT ( $p > 0.05$ ). NS = native starch; S1 = autoclaving-cooling 1 cycle; S2 = autoclaving-cooling 2 cycle; S3 = autoclaving-cooling 3 cycle; S4 = autoclaving-cooling 4 cycle; S5 = autoclaving-cooling 5 cycle.]

### 3.2. Proximate and Amylose Content of Starch

Table 1 demonstrates the proximate and amylose composition of both native starch and starch modified with autoclaving-cooling. The results indicated that the lipid content varied from 0.29-0.47% (db). Both the native starch and the starch modified by autoclaving-cooling showed no significant difference. Protein and moisture content were measured at 0.37-1.14% (db) and 9.06-11.39%, respectively. In comparison to the native starch, the protein and moisture content of the starch that was modified by autoclaving-cooling using 1, 2, 3, and 4 cycles decreased and significant difference. The ash content of native starch and autoclaving-cooling starch ranged from 0.22-2.92% (db), the ash content increased with the autoclaving-cooling modification. Native and autoclaving-cooling starch showed significant difference. Sweet potato starch exhibited increased protein content despite having ash and lipid levels remain comparable between native starch and autoclaved-cooled starch (Surenbra Babu & Parimalavalli, 2013). The autoclaving-cooling approach applied to cowpea starches yielded distinct results compared to this investigation, indicating that the ash level was like that of native starch (Ratnaningsih *et al.*, 2020).

Table 1. Proximate and amylose of native and modified starch

Parameter	NS	S1	S2	S3	S4	S5
Lipid (%db)	0.47±0.10 <sup>a</sup>	0.44±0.08 <sup>a</sup>	0.39±0.04 <sup>a</sup>	0.29±0.06 <sup>a</sup>	0.34±0.10 <sup>a</sup>	0.36±0.04 <sup>a</sup>
Protein (%db)	1.14±0.19 <sup>a</sup>	0.38±0.00 <sup>bc</sup>	0.37±0.01 <sup>c</sup>	0.47±0.12 <sup>bc</sup>	0.38±0.00 <sup>bc</sup>	0.81±0.35 <sup>ab</sup>
Ash (%db)	0.22±0.00 <sup>b</sup>	2.92±0.36 <sup>a</sup>	2.82±0.39 <sup>a</sup>	2.91±0.14 <sup>a</sup>	2.90±0.20 <sup>a</sup>	2.54±0.08 <sup>a</sup>
Moisture	11.39±0.50 <sup>a</sup>	10.52±0.40 <sup>bc</sup>	9.06±0.13 <sup>d</sup>	9.92±0.35 <sup>c</sup>	9.85±0.18 <sup>c</sup>	10.87±0.0 <sup>ab</sup>
Carbohydrate by difference	86.77±0.41 <sup>ab</sup>	85.75±0.84 <sup>bc</sup>	87.36±0.49 <sup>a</sup>	86.41±0.16 <sup>abc</sup>	86.54±0.47 <sup>abc</sup>	85.43±0.38 <sup>c</sup>
Amylose (%db)	18.58±0.10 <sup>d</sup>	21.94±0.07 <sup>bc</sup>	25.14±0.82 <sup>a</sup>	22.62±0.20 <sup>b</sup>	22.25±0.33 <sup>bc</sup>	21.57±0.18 <sup>c</sup>

Note: All values are presented as mean ± SD. Different superscript letters within the same row indicate significant differences based on DMRT ( $p < 0.05$ ). NS: native starch; S1: autoclaving-cooling 1 cycle; S2: autoclaving-cooling 2 cycle; S3: autoclaving-cooling 3 cycle; S4: autoclaving-cooling 4 cycle; S5: autoclaving-cooling 5 cycle.

The amylose content of native starch and autoclaving-cooling starch ranged from 18.58-25.14%. Native and autoclaving-cooling starch in all cycles showed significant difference in amylose content. All starches modified with autoclaving-cooling showed an increase in amylose content. The highest amylose content was observed after two cycles of autoclaving-cooling. The starch from *Tacca leontopetaloides* had the most amylose after 2 cycles of autoclaving-cooling, but the amylose amount decreased after 3 cycles (Herawati *et al.*, 2020). The *Tacca leontopetaloides* starch hydrolyzed by citric acid had 45.75% amylose, which increased to 59.90% following the addition of citric acid hydrolysis treatment to the autoclaving-cooling method. The process of using acids and enzymes before autoclaving aims to break the connections in the starch, leading to shorter chains of glucan. It results in an increase in amylose content (Nurhayati *et al.*, 2022). Retrograded starch exhibits increased amylose content due to partial degradation of amylopectin. This mechanism is related to the high pressure and temperature during the autoclave process (Ratnaningsih *et al.*, 2020). The amylose content in non-waxy rice starch reduces after one autoclaving-cooling cycle, remaining constant after three and five cycles when compared to native starch. In contrast, waxy rice starch subjected to one, three, and five autoclaving-cooling cycles demonstrates an amylose content like that of native starch (Sangwongchai *et al.*, 2024). Different amylose levels may be related to the type of material used or the modification process carried out.

### 3.3. Water Holding Capacity (WHC) and Oil Holding Capacity (OHC) of Starch

Figure 2 displays the water holding capacity (WHC) and oil holding capacity (OHC). The WHC values for native starch and starch modified with autoclaving-cooling range from 0.98 to 2.27 g/g. There was an increase in WHC value when comparing native starch to starch modified with autoclaving-cooling and showed significant difference. The WHC value is important in identifying specific uses. Many variables, such as source, modification, granule size, and starch structure, influence the WHC value (Makroo *et al.*, 2021). Autoclaving-cooling in one and two cycles increased WHC in comparison to that in five cycles. Ratnaningsih *et al.* (2020) observed similar findings, showing an increase in the WHC value from 0.69% for native starch to 3.04%, 3.06%, and 3.14% for modified starch during autoclaving-cooling in 1, 3, and 5 cycles, respectively. High amylose contents in starch exhibit significant water absorption ability,

affecting the WHC value (Rahmayuni *et al.*, 2024). The OHC value for native starch and starch modified by autoclaving-cooling was 1.09-1.42 g/g. The OHC value exhibited varying results between native and modified starch after autoclaving-cooling and showed significant difference. Autoclaving followed by cooling increased the OHC value of *Dioscorea esculenta* starch. White jack bean flour exhibited an increase in the OHC value from 106% to 116% following three autoclaving-cooling cycles (Rahmawati *et al.*, 2018). The investigation demonstrated that varied number of autoclaving-cooling cycles did not impact on the OHC value.

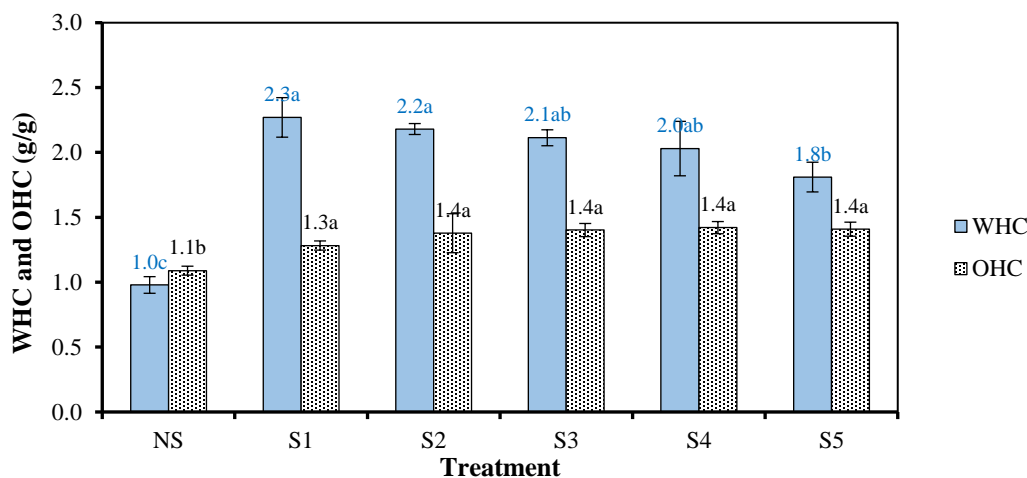


Figure 2. WHC and OHC of starch. [Note: All values are presented as mean  $\pm$  SD. Different letters in each parameter indicate significant differences based on DMRT ( $p < 0.05$ ). NS = native starch; S1 = autoclaving-cooling 1 cycle; S2 = autoclaving-cooling 2 cycle; S3 = autoclaving-cooling 3 cycle; S4 = autoclaving-cooling 4 cycle; S5 = autoclaving-cooling 5 cycle.]

### 3.4. Solubility and Swelling Power of Starch

Figures 3 and 4 show the solubility and swelling power at 60, 70, and 80 °C. An increase in temperature led to an increase in the solubility of both native and modified starch. At temperatures of 60, 70, and 80 °C, native and modified starches showed different solubility in all temperature. Similar results were shown in taro starch, where its solubility at a temperature of 65-95 °C increased after autoclaving-cooling (Setiarto *et al.*, 2020). Autoclaving-cooling on corn starch sample resulted in improved solubility values (Dundar & Gocmen, 2013). The combination of acid hydrolysis and autoclaving modifies starch and has the highest solubility. Similar results were shown in ginseng starch modified using citric acid-autoclaving treatment. This result is related to the presence of large amounts of short-chain amylose (Cui *et al.*, 2024). Increased solubility is associated with changes in structure, reduced molecular weight, and increased amylose (Raungrumsee & Anal, 2019). In this study, the increase in solubility value can be associated with an increase in amylose after modification was carried out.

Swelling power is a measure of water absorption in granules after heating (Khawas & Deka, 2017). The swelling power values of both native and modified starch, after autoclaving-cooling, exhibited an increase with higher temperatures. The swelling power increases at elevated temperatures due to enhanced gelatinization (Babu & Parimalavalli, 2013). Gelatinization causes damage to starch granules, resulting in irreversible changes in starch properties, such as granular swelling, loss of birefringence, and starch solubility. The temperature of the starch suspension is higher than its gelatinization temperature. A suspension temperature higher than the gelatinization temperature causes the breaking of hydrogen bonds. This breaking of hydrogen bonds results in water molecules being able to penetrate the starch granules and hydrate the free hydroxyl groups, resulting in starch swelling (Zavareze *et al.*, 2010). The swelling power value at 60 °C of the modified starch decreased compared to native starch. The swelling power at temperatures of 70 and 80°C showed no significant difference for both native and modified starch. The improved crystalline and strengthened interactions between amylose-amylose and amylose-amylopectin chains resulted in a reduction in swelling power. Modifications to the starch structure and the reassociation of starch chains after autoclaving improve swelling power (Xu *et al.*, 2023).

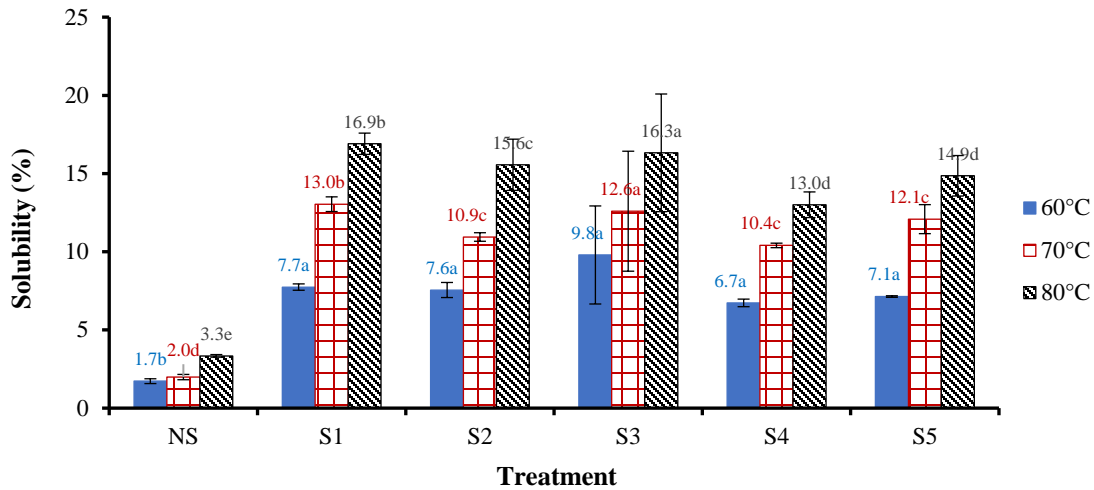


Figure 3. Solubility of starch. [Note: All values are presented as mean ± SD. Different superscript letters in each parameter indicate significant differences based on DMRT ( $p < 0.05$ ). NS = native starch; S1 = autoclaving-cooling 1 cycle; S2 = autoclaving-cooling 2 cycle; S3 = autoclaving-cooling 3 cycle; S4 = autoclaving-cooling 4 cycle; S5 = autoclaving-cooling 5 cycle.]

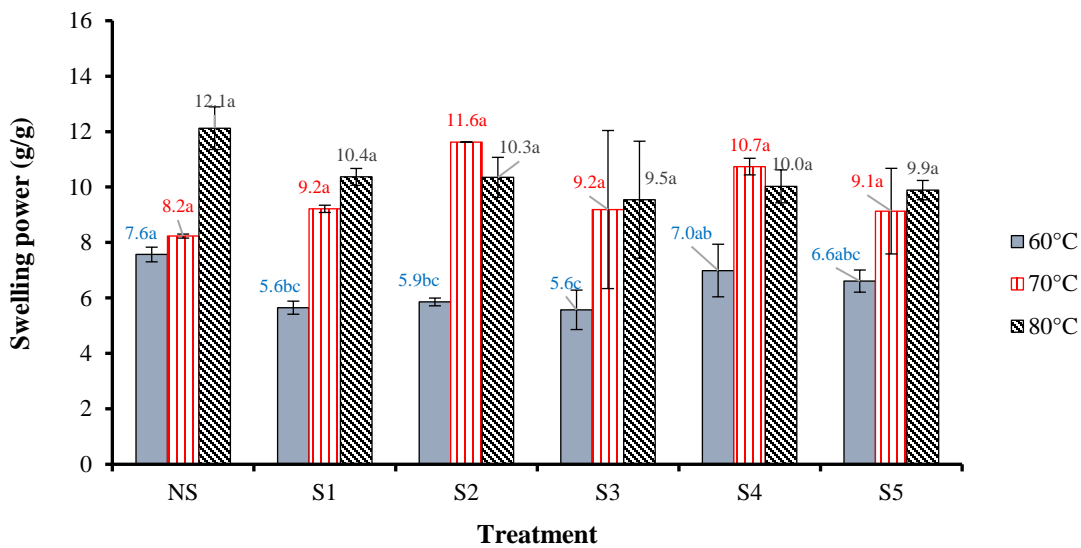


Figure 4. Swelling power of starch. [Note: All values are presented as mean ± SD. Different superscript letters in each parameter indicate significant differences based on DMRT ( $p < 0.05$ ). NS = native starch; S1 = autoclaving-cooling 1 cycle; S2 = autoclaving-cooling 2 cycle; S3 = autoclaving-cooling 3 cycle; S4 = autoclaving-cooling 4 cycle; S5 = autoclaving-cooling 5 cycle.]

### 3.5. Color of Starch

The color values of native and autoclaving-cooling starches are displayed in Table 2. The  $L^*$ ,  $a^*$ , and  $b^*$  values indicate lightness, redness, and yellowness, respectively (Ashwar *et al.*, 2016). The  $L^*$ ,  $a^*$ , and  $b^*$  values varied from 84.54 to 92.41; -6.58 to -3.14; and 9.12 to 14.84, respectively. The autoclaving-cooling method resulted in a decrease in the  $L^*$  value but an increase in the  $a^*$  and  $b^*$  values. The value showed significant difference between native and modified starch. The same result showed in arenga starch that modified with autoclaved (Ratnaningsih & Arovah, 2025). Cardaba banana starch was modified by heat-moisture treatment, resulting in a decrease in the  $L^*$  value and an increase in the  $a^*$  value (Olawoye *et al.*, 2022). Foxtail millet starch treated with autoclaving-cooling showed decreased  $L^*$  and  $a^*$  values, while  $b^*$  values increased (Surawan *et al.*, 2024).

Table 2. Color of native and modified starch

Parameter	NS	S1	S2	S3	S4	S5
L	92.41±0.11 <sup>a</sup>	86.35±0.85 <sup>b</sup>	86.29±0.41 <sup>b</sup>	84.87±1.83 <sup>b</sup>	85.30±0.47 <sup>b</sup>	84.54±0.91 <sup>b</sup>
a*	-6.58±0.32 <sup>c</sup>	-4.00±0.05 <sup>b</sup>	-3.74±0.41 <sup>ab</sup>	-3.14±0.26 <sup>a</sup>	-3.29±0.46 <sup>ab</sup>	-3.49±0.10 <sup>ab</sup>
b*	9.12±0.99 <sup>b</sup>	12.46±0.69 <sup>a</sup>	13.82±1.98 <sup>a</sup>	14.84±0.10 <sup>a</sup>	14.68±1.46 <sup>a</sup>	14.33±0.61 <sup>a</sup>
WI (%)	86.43±0.76 <sup>a</sup>	81.08±0.17 <sup>b</sup>	80.16±1.59 <sup>b</sup>	78.56±1.33 <sup>b</sup>	78.95±1.27 <sup>b</sup>	78.63±1.09 <sup>b</sup>
Chroma	11.25±0.99 <sup>b</sup>	13.09±0.64 <sup>ab</sup>	14.33±1.80 <sup>a</sup>	15.17±0.04 <sup>a</sup>	15.05±1.32 <sup>a</sup>	14.75±0.62 <sup>a</sup>
Hue	125.89±1.66 <sup>a</sup>	107.82±1.13 <sup>b</sup>	105.37±3.69 <sup>bc</sup>	101.96±1.02 <sup>c</sup>	102.75±2.92 <sup>bc</sup>	103.69±0.17 <sup>bc</sup>
ΔE	-	7.43±0.36 <sup>a</sup>	8.28±1.57 <sup>a</sup>	10.10±1.52 <sup>a</sup>	9.63±1.34 <sup>a</sup>	9.94±1.01 <sup>a</sup>

Note: All values are presented as mean ± SD. Different superscript letters within the same row indicate significant differences based on DMRT ( $p < 0.05$ ). NS: native starch; S1: autoclaving-cooling 1 cycle; S2: autoclaving-cooling 2 cycle; S3: autoclaving-cooling 3 cycle; S4: autoclaving-cooling 4 cycle; S5: autoclaving-cooling 5 cycle.

The color analyses show the values of the whiteness index (WI), chroma, hue, and total color difference (ΔE), the value is 78.56-86.43%; 11.25-15.17; 101.96-125.89; and 7.43-10.10, respectively. The WI value of modified starch decreased, and similar results were found in cowpea starch. It is suggested that the Maillard reaction during the autoclaving procedure contribute to the color change (Ratnaningsih *et al.*, 2020). The chroma values increased after the autoclaving-cooling procedure in all cycles. The same results were shown in foxtail millet starch after the autoclaving-cooling treatment. The higher the chroma value, the more intense the color (Surawan *et al.*, 2024). NS showed the highest hue angle, which significantly differed from starch after modification using autoclaving-cooling. The modification led to a decrease in hue angle, which coincided with an increase in the intensity of red and yellow colors (Sherin *et al.*, 2024).

There was no significant difference in ΔE values during all autoclaving-cooling cycles. Ačkar *et al.* (2010) categorized ΔE values into five distinct classifications: ΔE 0.5–1.5 as hardly apparent to the human eye, ΔE 1.5–3.0 as apparent to trained individuals, and ΔE 3–6 as being detectable by the general population. If ΔE is between 6 and 12, the color change is significant, although the color group remains. ΔE > 12 indicates that the samples are categorized into different color groups. The ΔE value of starch after modification by autoclaving-cooling was between 7.43 and 10.10, which indicates a significant color change classification.

#### 4. CONCLUSIONS

The study showed the effect of citric acid and autoclaving-cooling cycle treatments on the chemical and physical characteristics of *Dioscorea esculenta* starch. The results showed similar recovery yields, lipid, protein, moisture, and carbohydrate values between native starch and starch modified by autoclaving-cooling. The highest amylose content was observed after the starch modification in two cycles of autoclaving-cooling. The WHC value showed an increase after the autoclaving-cooling process, while the OHC value demonstrated similar results. The solubility and swelling power values exhibited an enhancement with increasing temperatures. Starch modified with autoclaving-cooling exhibited higher solubility than native starch, but swelling power demonstrated opposite results at a temperature of 60 °C. The L\* and WI values demonstrated a decrease following modification. The results of the current study recommend administering two autoclaving-cooling cycles of *Dioscorea esculenta* starch.

#### AUTHOR CONTRIBUTION STATEMENT

Author	C	M	So	Va	Fo	I	R	D	O	E	Vi	Su	P	Fu
AP	✓	✓	✓		✓	✓			✓	✓	✓			
YP	✓	✓		✓						✓	✓	✓		
PT	✓	✓		✓						✓	✓	✓		
DLNF	✓	✓		✓						✓	✓	✓		

C: Conceptualization	Fo: Formal Analysis	O: Writing - Original Draft	Fu: Funding Acquisition
M: Methodology	I: Investigation	E: Writing - Review & Editing	P: Project Administration
So: Software	D: Data Curation	Vi: Visualization	
Va: Validation	R: Resources	Su: Supervision	

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