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# The Effects of Heat-Moisture Modified Tapioca Starch on Dough Rheology and Bread Quality

Isı-Nem Uygulamasıyla Modifiye Edilmiş Tapyoka Nişastasının Hamur Reolojisi ve Ekmek Kalitesi Üzerine Etkileri

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#### Abstract

In this study, the effects of substituting wheat flour with 5% and 10% tapioca starch (TS) and heat-moisture modified tapioca starch (MTS) on dough rheology and bread quality were investigated. The addition of MTS increased the water absorption of wheat flour (p<0.05). While the use of TS and MTS reduced stability, they increased the degree of softening (p<0.05). The highest stability (13.50 min) and the lowest degree of softening (22.00 BU) were found in the control sample (p<0.05). While the addition of TS did not change the maximum resistance value of the flour (p>0.05), the addition of 10% MTS decreased this value (p<0.05). The addition of MTS increased the moisture content of bread samples (p<0.05). No significant difference was determined among the specific volume values of the samples (p>0.05). With the addition of TS,  $L^*$  and  $b^*$  values in bread crusts increased, while the *a*\* value decreased (p<0.05). The crust color values of bread samples containing 10% MTS were found to be similar to the control sample (p>0.05). With the addition of TS, the number of pores decreased, and the area increased, resulting in fewer but larger pores (p<0.05). The pore structure and textural properties of bread samples produced with the addition of 5% MTS was found to be similar to the control sample (p>0.05). 5% of MTS can be used as a substitute for wheat flour in bread production.

**Keywords:** Bread; Extensograph; Farinograph; Pore structure; Tapioca starch; Texture

# 1. Introduction

Bread is an important baked product because of its high consumption. Wheat flour is the main ingredient for bread production. However, due to the factors such as high demand, geographical conditions, climate change, and inadequate functionality, alternative sources are being sought. The quality of bread made from wheat flour cannot be achieved with substitutes that do not contain gluten. Therefore, improving the technological properties of substances that will be substituted for wheat flour is of great importance (Rodriguez-Sandoval *et al.* 2017).

# Öz

Bu çalışmada, buğday ununun %5 ve %10 oranında tapyoka nişastası (TS) ve ısı-nem uygulamasıyla modifiye edilmiş tapyoka nişastasıyla (MTS) ikamesinin hamur reolojisi ve ekmek kalitesi üzerine etkileri incelenmiştir. MTS ilavesi, buğday ununun su absorbsiyonunu artırmıştır (p<0.05). TS ve MTS kullanımı stabiliteyi azaltırken, yumuşama derecesini artırmıştır (p<0.05). En yüksek stabilite (13.50 dakika) ve en düşük yumuşama derecesi (22.00 BU) kontrol örneğine ait bulunmuştur (p<0.05). TS ilavesi buğday ununun maksimum direnç değerini değiştirmemiştir (p>0.05), ancak %10 MTS ilavesi bu değeri azaltmıştır (p<0.05). MTS ilavesi ekmek örneklerinin nem içeriğini artırmıştır (p<0.05). Örneklerin spesifik hacim değerleri arasında önemli bir fark belirlenmemiştir (p>0.05). TS ilavesiyle ekmek kabuklarının  $L^*$  ve  $b^*$  değerleri artarken,  $a^*$  değeri azalmıştır (p<0.05). %10 MTS içeren ekmek örneklerinin kabuk renk değerleri kontrol örneği ile benzer bulunmuştur (p>0.05). TS ilavesiyle gözenek sayısı azalırken toplam gözenek alanı artmıştır, dolayısıyla daha az sayıda ve daha büyük gözenekler oluşmuştur (p<0.05). %5 MTS ilavesiyle üretilen ekmek örneklerinin gözenek yapısı ve tekstürel özellikleri kontrol örneğiyle benzer bulunmuştur (p>0.05). Buğday ununa %5 MTS ikame edilerek ekmek üretiminde kullanılabileceği sonucuna varılmıştır.

Anahtar Kelimeler : Ekmek; Ekstensograf; Farinograf; Gözenek yapısı; Tapyoka nişastası; Tekstür

Cassava is a plant that has low nutritional requirements and is also drought resistant (Milde *et al.* 2012). Therefore, it is a good alternative to carbohydrate sources that require specific soil and climate conditions. Kızıl-Aydemir *et al.* (2019) reported that the cassava plant is an important energy crop that can be cultivated in arid and infertile lands in Türkiye. While cassava refers to the root of the plant, tapioca is used to describe the starch and other processed products obtained from this plant (Hsieh *et al.* 2019). Tapioca starch can be derived from cassava root and has appealing properties for food formulations (Kaveh *et al.* 2020). The isolation of starch from cassava tubers is relatively easier than from other starchy plants due to its low protein, fat, and fiber content (Javadian et al. 2021). Tapioca starch is hypoallergenic and contains high molecular weight amylose. It can develop a less firm gel due to its lower amylose content, and it has a lower gelatinization temperature compared to many other starches (Kaveh et al. 2020). Tapioca starch is low-priced, has high viscosity, a mild flavor, and a clear paste appearance (Javadian et al. 2021). Tapioca starch is particularly a good alternative for products where wheat flour is imported and some studies showed its substitution for wheat flour. However, the use of tapioca starch in bread production has disadvantages due to the absence of gluten, such as increase in hardness, decrease in specific volume, dense texture and heterogeneous pore structure (Prameswari et al. 2018).

The modification can improve the technological properties of starch such as; the advanced cooking characteristics, improved texture, increase in freeze-thaw stability and delay of staling (Miyazaki *et al.* 2008). Additionally, through modification, the properties of starch such as solubility, microstructure, adhesion, gel clarity, film-forming ability, gel transparency, gel tendency, and syneresis can also be altered and improved (Javadian *et al.* 2021).

The physical, chemical, or enzymatic treatments can be used for modification of starch, thus the molecular organization of starch can change or be rearranged, polymers can be degraded, or incorporation of chemical groups can be allowed via modification (Dariva et al. 2021). Among these methods, the physical modification method is widely used because it is not a chemical process, it is an environmentally friendly method, and it does not result in the formation of harmful by-products. Physical modification can be applied through thermal and non-thermal methods. The hydrothermal modification method is a simple, easy, safe, low-cost, and green physical modification method. In this method, starch is heated above the gelatinization temperature at limited moisture content (10-35%). The process can be applied for 15 minutes to 16 hours (Marta et al. 2022).

The method performed at high humidity and below the gelatinization temperature but above the glass transition temperature is a modification that can improve the physicochemical properties of starch without altering its granular structure. This process can modify the starch's molecular weight (amylose/amylopectin ratio), microscopic crystalline structure, physicochemical properties, and in vitro digestibility (Yalçın *et al.* 2020). Modified starches can be incorporated into bread

formulations by substituting wheat flour for various purposes, such as improving product quality and delaying staling (Rodriguez-Sandoval *et al.* 2017).

In this study, the potential use of modified tapioca starch which has been modified using a hydrothermal method in bread production has been investigated. Wheat flour was substituted with both tapioca starch and modified tapioca starch to investigate the rheological properties of the flour. Additionally, the aim of this study was to determine the effect of modified tapioca starch on the quality characteristics of bread.

### 2. Materials and Methods

### 2.1 Materials

Tapioca starch (TS), wheat flour (WF), salt, sugar and yeast were purchased from commercial companies in Konya, Türkiye.

# **2.2** Modification of tapioca starch by heat-moisture treatment

The modified tapioca starch (MTS) was prepared using the method of Marta *et al.* (2022). The moisture content of TS was adjusted to 30%, and then the starch was kept into an air-tight container at 4°C for 24 h. The starch taken out of the refrigerator was kept in an oven at 100°C for 16 hours. Afterward, it was dried under 10% moisture content at 50°C for 24 hours, ground, and sieved through a 150-micron sieve. The heat-moisture treated TS was obtained and kept at room temperature until further use.

# 2.3 Bread production

Bread production was carried out according to AACC (1999) and Saka et al. (2021). WF was substituted with TS or MTS at different ratios (0, 5 and 10%). To make dough, 1% sugar, 1.5% salt, and 3% yeast were added to the flour, and water was added depending on the amount determined in the farinograph. The mixture was kneaded for 10 minutes in a kitchen-type dough kneader (KitchenAid, 5KSM45, USA) at a slow speed. The kneaded dough was allowed to rest at room temperature for 30 min. Subsequently, 70 grams of dough were hand-shaped into rounds and proofed at 30°C and 80% relative humidity for 30 min. After fermentation, the dough samples were baked at 250°C for 10 minutes in an electrical oven (Model HN678G4S6, Siemens, Munich, Germany). The breads samples are shown in Figure 1. The control bread was produced with 100% WF. Bread variants containing 5% TS, 10% TS, 5% MTS and 10% MTS were referred as 5TS, 10TS, 5MTS and 10MTS, respectively.

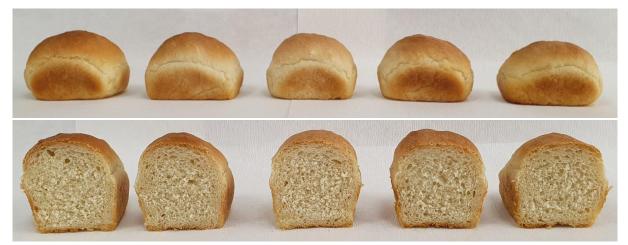


Figure 1. The bread samples (from left to right, control, 5TS, 10TS, 5MTS, 10MTS bread samples)

#### 2.4 Rheological analyses

The rheological characteristics of flour and starch mixes were assessed using the farinograph (Brabender GmbH & Co KG, Germany) according to Approved Method 54-21, and the extensograph (Brabender GmbH & Co KG, Germany) according to Approved Method 54-10 (AACC 2010). Only the values at 135 min among the extensograph parameters were discussed.

# **2.5** Determination of moisture content and specific volume

The moisture content and specific volume values of bread samples were determined using the methods outlined in AACC Standard Method No: 44-01.01 and AACC Standard Method No: 10-05.01, respectively (AACC, 2010).

#### 2.6 Color measurement

The *L*\*, *a*\*, and *b*\* color parameters of both the crumb and crust of bread samples were measured using a Minolta CR300 (Minolta Inc., Tokyo, Japan).

# 2.7 Crumb pore count and total pore area measurement

The crumb pore count and total pore area of bread samples were measured using ImageJ software according to the method described by our previous study (Çetin-Babaoğlu *et al.* 2023). A flatbed scanner (HP LaserJet M1120 MFP, USA) was used to scan the bread slices. First, the unit of length was converted from pixels to cm. The image, converted to grayscale format, was masked using thresholding and transformed into a binary image. Finally, data were obtained through particle analysis.

# 2.8 Texture profile analysis

The texture parameters (hardness, springiness, cohesiveness, chewiness) of bread samples was analysed using by a texture analyser (TA-TX2i, Stable Micro System, Surrey, UK) according to the method described by Zhou et al. (2021). A 2-cm thick slice was taken from the centre of

the samples with a bread knife. A 30-mm diameter cylindrical probe was used. The compression rate, the test speed, the pre-test speed and the post-test speed were 40%, 1 mm/s, 1.7 mm/s and 1 mm/s, respectively.

# 2.9 Statistical analysis

The experiments were replicated twice. All data, presented as the mean of at least three measurements, were statistically analyzed using Minitab 16 software (Minitab Inc., USA) through one-way analysis of variance (ANOVA). Significant differences among group means were determined using Tukey's test (p<0.05)

#### 3. Results and Discussions

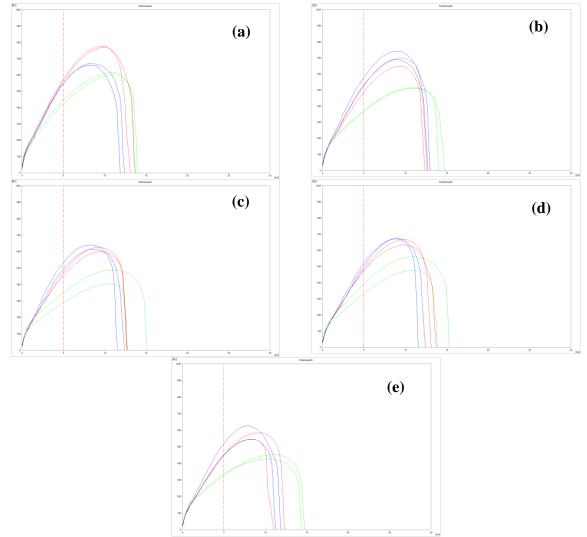
# 3.1 Rheological properties of flour blends

The rheological behaviors of all flour blends during the farinograph and extensograph tests are provided in Table 1. Additionally, the extensograph graphs at 45, 90, and 135 minutes are illustrated in Figure 2. Although the water absorption of wheat flour did not change with the addition of TS (p>0.05), the addition of MTS increased water absorption (p<0.05). The highest water absorption value belonged to the sample containing 10% MTS (p<0.05). The sample with the shortest development time was the one containing 10% TS (p<0.05). The development time of other samples was found to be similar to the control sample (p>0.05). While the addition of TS and MTS reduced the stability value of wheat flour, it increased the degree of softening (p<0.05). Upon examination of the extensograph results, no significant disparity was observed among the energy, resistance to extension, and extensibility values of flour blends (p>0.05). However, the addition of 10% TS and 10% MTS notably diminished the maximum resistance value of wheat flour (p<0.05). The sample containing 10% MTS exhibited the lowest maximum resistance value (p<0.05).

Table 1. The farinogram and extensogram (at 135 min proofing time) characteristics of wheat flour and mixtures containing wheat
flour and tapioca starch or modified tapioca starch in various proportions (mean $\pm$ std error)

Flour mixture	Water Absorption (%)	Development Time (dk)	Stability (dk)	Degree of Softening (BU)
WF	59.90 ± 0.10 <sup>c</sup>	2.30 ± 0.00 <sup>a</sup>	13.50 ± 0.95 <sup>a</sup>	22.00 ± 2.01 <sup>b</sup>
95WF:5TS	59.80 ± 0.00 <sup>c</sup>	$1.80 \pm 0.10^{ab}$	$2.40 \pm 0.20^{b}$	$54.00 \pm 4.01^{a}$
90WF:10TS	$60.00 \pm 0.20^{\circ}$	$1.50 \pm 0.00^{b}$	$2.00 \pm 0.10^{b}$	$57.00 \pm 1.00^{a}$
95WF:5MTS	62.80 ± 0.05 <sup>b</sup>	$2.30 \pm 0.18^{a}$	$2.60 \pm 0.20^{b}$	$51.80 \pm 2.26^{a}$
90WF:10MTS	$64.80 \pm 0.10^{a}$	$1.90 \pm 0.00^{ab}$	2.20 ± 0.05 <sup>b</sup>	53.80 ± 2.76 <sup>a</sup>
Flour mixture	Energy (A)	Resistance to Extension	Extensibility (E)	Maximum Resistance
	(cm²)	(R50) (BU)	(mm)	(Rm) (BU)
WF	$104.00 \pm 4.01^{a}$	552.00 ± 30.41 <sup>a</sup>	122.00 ± 10.53ª	665.00 ± 31.59 <sup>ab</sup>
95WF:5TS	116.00 ± 7.02 <sup>a</sup>	557.00 ± 13.04 <sup>a</sup>	129.00 ± 8.02 <sup>a</sup>	716.00 ± 14.04 <sup>a</sup>
90WF:10TS	96.00 ± 9.03 <sup>a</sup>	514.00 ± 14.04 <sup>a</sup>	120.00 ± 4.01 <sup>a</sup>	628.00 ± 7.02 <sup>bc</sup>
95WF:5MTS	100.00 ± 8.02 <sup>a</sup>	514.00 ± 20.06 <sup>a</sup>	120.00 ± 9.03 <sup>a</sup>	674.00 ± 19.06 <sup>ab</sup>
90WF:10MTS	86.00 ± 8.53 <sup>a</sup>	488.00 ± 22.07 <sup>a</sup>	$116.00 \pm 5.01^{a}$	586.00 ± 24.07 <sup>c</sup>

Superscript letters in the same column means statistical significance (p<0.05). WF: 100% wheat flour; 95WF:5TS: 95% wheat flour + 5% tapicca starch; 90WF:10TS: 90% wheat flour + 10% tapicca starch; 95WF:5MTS: 95% wheat flour + 5% modified tapicca starch; 90WF:10MTS: 90% wheat flour + 10% modified tapicca starch.



**Figure 2.** The extensogram characteristics of flour mixtures at 45 (green line), 90 (red line), 135 (blue line) min (The graphics are belonged to a) WF, b) WF with 5% TS, c) WF with 10% TS, d) WF with 5% MTS e) WF with 10% MTS)

The rheological characteristics of flour are mostly dependent on protein content and quality. The reduction in gluten content can led to rapid development and hydration. Thus the development time and stability decrease, while the degree of softening increases (Mohebbi *et al.* 2018). Additionally, as energy, resistance to extension, and maximum resistance values decrease,

extensibility value increases (Çetin-Babaoğlu *et al.* 2022). The rheological values obtained from this study could be resulted from dilution of gluten with the addition of TS or MTS. However, the addition of MTS positively influenced the water absorption value of the flour. The modification process causes the disruption of hydrogen bonds between the amorphous and crystalline regions of starch,

leading to the expansion of the amorphous region. As a result, the hydrophilic property of starch increases. Additionally, the starch granule surface becomes more porous. For these reasons, the water absorption capacity of modified starch is higher than that of native starch. Additionally, the rearrangement of molecules, changes in gel strength, formation of cross-links, and reduction in the degree of double helix as a result of modification in starch may have also influenced the rheological properties. (Marta et al. 2022). Miyazaki et al. (2008) reported that the substituted samples which tend to shorten development time, show higher amount of water absorption. They stated that this result is relate hydrophilic property of starch. They determined, unlike this study, that the addition of TS and MTS reduced the water absorption of wheat flour. However, the starch

Table 2. The physical characteristics of bread samples

modification method used is crucial on these properties and the heat-moisture treatment method used in this study increased the water absorption value of TS.

# **3.2** Moisture contents and specific volumes of bread samples

The moisture content and specific volume values of bread samples are shown in Table 2. The addition of 10% MTS increased the moisture content of the bread sample (p<0.05). However, the moisture contents of the other bread samples were found to be similar to the control group (p>0.05). The addition of TS and MTS did not cause a significant change in specific volume of the bread samples (p>0.05).

Analyses		Control	5TS	10TS	5MTS	10MTS
Moisture of	content (%)	41.44 ± 0.30 <sup>b</sup>	40.84 ± 0.09 <sup>b</sup>	41.30 ± 0.12 <sup>b</sup>	41.84 ± 0.42 <sup>ab</sup>	43.03 ± 0.12 <sup>a</sup>
Specific vo (ml/g)	lume	2.85 ± 0.05 <sup>a</sup>	2.79 ± 0.04 <sup>a</sup>	2.79 ± 0.01ª	2.82 ± 0.00 <sup>a</sup>	$2.71 \pm 0.04^{a}$
	L*	61.00 ± 0.31 <sup>c</sup>	65.74 ± 0.20 <sup>ab</sup>	68.92 ± 1.18 <sup>a</sup>	65.20 ± 0.36 <sup>b</sup>	63.52 ± 0.18 <sup>bc</sup>
CRUST	a*	13.09 ± 0.34 <sup>a</sup>	11.09 ± 0.52 <sup>ab</sup>	9.65 ± 0.36 <sup>b</sup>	11.51 ± 0.39 <sup>ab</sup>	$12.30 \pm 0.20^{a}$
	b*	32.67 ± 0.12 <sup>c</sup>	34.34 ± 0.45 <sup>ab</sup>	35.34 ± 0.24 <sup>a</sup>	34.19 ± 0.07 <sup>ab</sup>	33.00 ± 0.12 <sup>bc</sup>
	ΔE	-	5.43 ± 0.50 <sup>ab</sup>	$9.04 \pm 0.24^{a}$	4.77 ± 0.21 <sup>b</sup>	2.67 ± 0.12 <sup>b</sup>
	L*	76.19 ± 0.95 <sup>b</sup>	77.55 ± 0.35 <sup>ab</sup>	79.84 ± 0.62 <sup>a</sup>	77.42 ± 0.04 <sup>ab</sup>	78.61 ± 0.20 <sup>ab</sup>
CRUMB	a*	-2.14 ± 0.09 <sup>a</sup>	-2.14 ± 0.04 <sup>a</sup>	$-2.30 \pm 0.04^{a}$	-2.24 ± 0.05 <sup>a</sup>	-2.25 ± 0.02 <sup>a</sup>
	b*	21.11 ± 0.28 <sup>a</sup>	21.45 ± 0.12 <sup>a</sup>	$21.21 \pm 0.72^{a}$	21.57 ± 0.17 <sup>a</sup>	21.17 ± 0.01 <sup>a</sup>
	ΔE	-	1.42 ± 0.31 <sup>b</sup>	3.73 ± 0.63 <sup>a</sup>	$1.32 \pm 0.09^{b}$	$2.42 \pm 0.19^{ab}$
Pore coun	t/cm²	18.38 ± 0.38 <sup>a</sup>	14.13 ± 0.38 <sup>bc</sup>	12.63 ± 0.88 <sup>c</sup>	16.50 ± 0.50 <sup>ab</sup>	14.50 ± 0.75 <sup>bc</sup>
Total pore area/cm <sup>2</sup>		0.47 ± 0.03 <sup>b</sup>	0.57 ± 0.00 <sup>ab</sup>	0.57 ± 0.00 <sup>a</sup>	0.48 ± 0.01 <sup>ab</sup>	0.52 ± 0.03 <sup>ab</sup>

Superscript letters in the same line means statistical significance (p<0.05). Mean  $\pm$  std error (p <0.05).

Control: 100% wheat flour; 5TS: 5% tapioca starch + 95% wheat flour; 10TS: 10% tapioca starch + 90% wheat flour; 5MTS: 5% modified tapioca starch + 95% wheat flour; 10MTS: 10% modified tapioca starch + 90% wheat flour.

According to the farinograph results, since the addition of MTS increased the water absorption of the flour, it is expected that the moisture content of the bread samples containing MTS were high. Tapioca starch contains high levels of amylopectin (75-80%), which gives it a high swelling capacity (Hsieh et al. 201, Kaveh et al. 2020). This property can be further enhanced through modification methods. Manchun et al. (2012) reported that the swelling power of tapioca starch increased after modification with heat and ultrasound treatment. Furthermore, the water absorption capacity and solubility of starch granules are factors affecting swelling capacity and there is a positive relationship between them (Manchun et al. 2012). The increase in water absorption and swelling power can enhance the bread volume. With sufficient kneading, gluten can absorb an adequate amount of water and form a strong gluten matrix. The water and gas retained by the gluten matrix allow the dough to expand. Specific volume is inversely proportional to the weight of the bread and directly proportional to the bread volume. Excess water retained

by the dough can increase the specific volume (Kaveh et al. 2020). However, the amount and strength of gluten are important here. Adding a different substance to the flour can reduce the gluten concentration, leading to insufficient structure to retain the gas formed, and thus, a decrease in volume (Çetin-Babaoğlu et al. 2021). Despite not containing gluten, tapioca starch did not lead to a significant change in the specific volume values of bread samples in this study when substituted for wheat flour, both in its unprocessed form and after being modified by heat-moisture treatment. This is due to the complex mucoadhesive properties of tapioca starch, such as gelatinization and adhesiveness (Kaveh et al. 2020). In this study, the results showed that modification could enhance the technological properties of tapioca starch since it has the potential to enhance the dough's viscoelastic properties, allowing it to effectively capture and hold carbon dioxide gas bubbles generated during fermentation. It can provide cohesiveness, viscosity, and retain air bubbles in dough, in this way (Pongjaruvat et al. 2014).

### 3.3 Color values of bread samples

The color values of bread samples are given in Table 2. The addition of unprocessed or modified tapioca starch to bread increased the crust's brightness ( $L^*$ ) value (p<0.05). The lowest L\* value (61.00) was found in the control sample (p<0.05). The L\* value of the 10MTS sample's crust was similar to that of the control sample (p>0.05). The heat-moisture modification applied to the tapioca starch resulted in a decrease in the  $L^*$  value (p<0.05). In the 10TS crust sample, the brightness increased along with a decrease in the redness  $(a^*)$  value and an increase in the yellowness ( $b^*$ ) value (p<0.05). The lowest  $a^*$  value (9.65) belonged to the 10TS crust (p<0.05). The  $a^*$  values of the crust in the other samples were found to be similar to that of the control sample (p>0.05). The addition of unprocessed or modified tapioca starch to the bread increased the yellowness value of the crust samples (p<0.05). For all color parameters of the crust, the values for 10MTS were found to be similar to those of the control sample, with the differences being statistically insignificant (p>0.05). Looking at the crumb color values, the brightness value of the bread sample containing 10% TS (79.84) was the highest (p<0.05), while the L\* values of the other samples were similar to that of the control sample (p>0.05). The redness and yellowness values of the crumbs did not change significantly with the addition of unprocessed or modified tapioca starch (p>0.05). Looking at the  $\Delta E$  values, it is observed that the bread sample containing 10% TS exhibited the greatest color change compared to the control sample, both in the crust and crumb (p<0.05). The color change in the other samples was found to be similar to each other (p>0.05).

The color of bread is a key attribute, along with its texture and aroma, that significantly influences consumer preferences. This characteristic is influenced by various factors including the moisture content, pH level, reducing sugar concentration, and amino acid composition of the dough. Additionally, baking conditions such as temperature, relative humidity, and the method of heating also play a crucial role (Kim et al. 2015). Kim et al. (2015) reported that in their study of gluten-free bread produced by substituting rice flour with varying proportions of tapioca starch, the crust L\* value of the control sample was 66.29. However, in samples containing tapioca starch, the crust L\* value ranged between 66.66 and 68.38. Furthermore, similar to our results, they found a decrease in the crust's redness value  $(a^*)$  with the addition of tapioca starch. The yellowness value (b\*) of the bread crumb increased with the addition of tapioca starch. The mentioned data supports the findings of our study. Additionally, based on the obtained results, it can be said that the applied heat-moisture modification method has been effective on the color values of tapioca starch. The brightness value of tapioca

starch decreased, while the redness value increased and the yellowness value decreased due to the heat treatment. Thus, similar color values to those of the control sample were obtained.

# 3.4 Crumb pore count and total pore area measurement of bread samples

The crumb pore count and total pore area values of bread samples are given in Table 2. The addition of tapioca starch decreased the number of pores per unit area in the bread samples while increasing the total pore area (p<0.05). This resulted in a bread crumb with larger, more irregular pores. However, modified tapioca starch at a 5% addition rate gave results similar to the control sample (p>0.05). For bread samples containing 10% modified tapioca starch, the number of pores per unit area decreased (p<0.05), while the total pore area was similar to that of the control sample (p>0.05). The bread sample containing 10% tapioca starch had the highest total pore area and the lowest number of pores (p<0.05).

Pores are classified into three categories based on their sizes. Those with areas smaller than 4 mm<sup>2</sup> are categorized as small, those with areas ranging from 4-8 mm<sup>2</sup> are categorized as medium, and those with areas larger than 8 mm<sup>2</sup> are categorized as large pores. Pores smaller than 1 mm<sup>2</sup> are considered artifacts (Polaki et al. 2010). During the mixing of dough ingredients, air is trapped in the liquid phase of the dough in the form of small nuclei. Additionally, carbon dioxide formation occurs during fermentation. The subsequent dough processing stages allow for the division of gas cells, increasing their number and improving their size distribution. During baking, the gases expand, causing the gas nuclei to enlarge. Wheat flour dough has the ability to stabilize expanding gas cells due to its unique visco-elastic properties. The starch-gluten matrix is crucial for the stabilization of gas cells. As the baking process continues, the structure stabilizes through starch gelatinization, the formation of retrograded amylose aggregate, and protein denaturation, allowing the gas cells to remain stable (Sroan et al. 2009). Adding a gluten-free substance to wheat flour dilutes gluten, resulting in a coarse dough structure. This weakened structure struggles to adequately contain the gas generated during fermentation, leading to an inclination for expanding pores to merge, thereby reducing pore count and forming large, irregular pores (Çetin-Babaoğlu et al. 2023). Starch gelatinization plays a crucial role in improving the physical properties of bread in the absence of gluten and in enhancing the dough's gas retention capacity due to starch's ability to form a matrix that can trap gas. Gelforming starches can stabilize gas cells. In particular, starches such as corn, potato, rice, and tapioca are used for this purpose in gluten-free formulations (Horstmann et al. 2017). The modification process also affects starch's gelatinization, retrogradation properties, reactivity, and molecular arrangement. Consequently, it can influence the resulting starch matrix and gluten-starch interaction, thereby improving the stabilization of gas cells (Miyazaki *et al.* 2008; Dariva *et al.* 2021).

#### 3.5 Textural properties of bread samples

The values for the hardness, springiness, cohesiveness, and chewiness parameters from the texture profile analysis of bread samples are given in Table 3. The substitution of tapioca starch and modified tapioca starch did not have a significant effect on the springiness and chewiness values of the bread samples (p>0.05). However, tapioca starch increased the hardness value at both usage rates and decreased the cohesiveness value when used at a rate of 10% (p<0.05). The hardness and cohesiveness values of the bread samples with modified tapioca starch were found to be similar to those of the control sample (p>0.05). The decrease in gluten concentration in the dough leads to the disruption of the gluten network and results in an inadequate structure to retain the gas formed in the dough (Zhang *et al.* 2022). Consequently, the bread volume is low, and the bread crumb becomes denser and firmer. The pore structure is also a significant factor affecting textural properties. An increase in cohesiveness is an indicator of increased product volume and gas retention capacity (Marta *et al.* 2024).

**Table 3.** Texture profile parameters of bread samples

Bread Sample	Hardness (g)	Springiness	Cohesiveness	Chewiness
Control	3394.87 ± 43.52 <sup>b</sup>	0.97 ± 0.00 <sup>a</sup>	$0.61 \pm 0.01^{a}$	2030.66 ± 20.96 <sup>a</sup>
5TS	4178.73 ± 35.96 <sup>a</sup>	$0.95 \pm 0.00^{a}$	0.56 ± 0.01 <sup>ab</sup>	2120.80 ± 38.07 <sup>a</sup>
10TS	4238.72 ± 99.64 <sup>a</sup>	$0.94 \pm 0.00^{a}$	$0.54 \pm 0.01^{b}$	2252.93 ± 79.88 <sup>a</sup>
5MTS	3676.43 ± 22.06 <sup>ab</sup>	$0.96 \pm 0.01^{a}$	0.58 ± 0.01 <sup>ab</sup>	2053.22 ± 33.11 <sup>a</sup>
10MTS	3774.35 ± 269.71 <sup>ab</sup>	$0.96 \pm 0.00^{a}$	0.59 ± 0.01 <sup>ab</sup>	2112.52 ± 109.38 <sup>a</sup>

Superscript letters in the same column means statistical significance (p<0.05). Mean ± std error (p<0.05). Control: 100% wheat flour; 5TS: 5% tapioca starch + 95% wheat flour; 10TS: 10% tapioca starch + 90% wheat flour; 5MTS: 5% modified tapioca starch + 95% wheat flour; 10MTS: 10% modified tapioca starch + 90% wheat flour.

Since tapioca starch substitution reduces gluten concentration, it leads to an increase in bread hardness and a decrease in cohesiveness. However, the effects of tapioca starch, whose technological properties were improved through modification, on the textural properties of the bread were found to be statistically insignificant (p<0.05). Therefore, substituting modified tapioca starch wheat flour would be more suitable than native tapioca starch. Rodriguez-Sandoval et al. (2017) investigated the textural properties of bread samples produced by substituting wheat flour with modified cassava starch at different levels (5, 7, and 10%). According to their results, the use of modified cassava starch increased the hardness values of the samples. The cohesiveness values of all samples were found to be similar. The use of 10% cassava starch led to an increase in firmness values. They concluded that the use of 5% modified cassava starch was suitable in terms of texture parameters. Of course, the modification method applied is highly decisive for the technological properties of the starch. The study conducted by Wang et al. (2024) demonstrates the importance of modification methods on the technological properties of starch. In the mentioned study, it was found that dually modified quinoa starch resulted in bread samples with textural properties similar to the control sample, while starches modified by a single method reduced the hardness of the bread samples. A similar trend was observed in chewiness values. The modification of starch did not cause significant changes in the cohesiveness values of the

bread samples. However, compared to native starch, modified starch reduced the gumminess values of the bread samples. Considering all the obtained results, Wang et al. (2024) reported that modified starch contributes to the dough's gas retention capacity by forming a threedimensional gel structure in the dough, thereby reducing hardness, chewiness, and gumminess values. This situation may be related to the modification process altering the molecular arrangement of the starch, allowing for the formation of a stronger gel. As a result, the amount of water retained in the structure increases. Additionally, modification could lead to an increase in the amylose content, while expanding the amorphous regions, reducing the degree of double helices, and exposing more hydrophilic parts, which contribute to enhanced water absorption (Marta et al. 2022).

#### 4. Conclusions

Tapioca starch is an ingredient used in bread formulations. However, because it lacks gluten, substituting it for wheat flour can reduce bread quality. It is typically used as an alternative or supplement to hydrocolloids in gluten-free formulations due to its contribution to structure. Its gel-forming ability is particularly important for forming the structure of the product. Enhancing this property and water absorption capacity through various modification methods can improve bread quality, thus in breads made with wheat flour, it may be preferred for its contribution to shelf life or bread texture. This study found that heat-moisture modification improved some properties of native tapioca starch and concluded that using 5% modified tapioca starch in bread formulations yielded results similar to the control sample. The use of alternative flours and starch derivatives to wheat flour will increase agricultural diversity, and the widespread cultivation of crops with low climate and soil selectivity will contribute to the country's agriculture. Therefore, studies on the use of different materials in the production of bread, which is widely consumed, should be increased, and these literature data should be utilized for making the right choices. The cassava plant, being drought-resistant and having low soil and climate selectivity, has the potential to be cultivated in our country in the future. Therefore, flour and starch derived from this plant may be widely used in bakery products. Increasing production will also reduce the currently high cost. If high yields are achieved, the substitution rate of tapioca starch for wheat flour can be increased without compromising product quality. It would be useful to explore the application of different modification methods for this purpose. Although hydrothermal modification does not significantly increase the usage rate of tapioca starch as expected, the data obtained from this study will contribute to the literature. In future studies, the effects of starches whose properties, such as gel-forming ability, water absorption capacity, and retrogradation characteristics, are modified and improved through different modification methods on both bread quality and shelf life can be investigated.

#### **Declaration of Ethical Standards**

The authors declare that they comply with all ethical standards.

#### Credit Authorship Contribution Statement

Author- Conceptualization, Methodology / Study design, Formal analysis, Investigation, Resources, Data curation, Writing – original draft, Writing – review and editing.

#### **Declaration of Competing Interest**

The author declares no conflict of interest

# Data Availability

Not Applicable

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