



## The Effect of Microbial Transglutaminase (MTGase) Enzyme on Physical, Sensorial and Nutritional Properties of Atlantic Salmon (*Salmo salar* Linnaeus, 1758) Meatballs

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**Abstract:** In this research, fish balls were produced from Atlantic salmon (*Salmo salar* Linnaeus, 1758) with the addition of MTGase (0.60%), which is included in the seafood menus of catering companies, hotels, restaurants, and other ready-made food establishments in our country and especially around the world. In order not to risk food safety for such businesses where time and hygiene are very important, nutritional, physical, and sensory analyzes were carried out in fish balls to investigate the adequacy/applicability of the combined effect of short time and low temperature (4±1°C; 3 hours) for MTGase enzyme activation. After the production and activation processes, the meatballs were baked in the oven at 180 °C for 20 minutes. While the crude protein and moisture content of MTGase added group (B) was higher than the control (A) group (p<0.05), the energy content of group A was higher than group B (p<0.05). The pH values of groups A and B were determined as 7.40 and 7.32, respectively (p<0.05). It was also revealed that the brightness (L\*) value increased with the use of MTGase. MTGase activation of short-term and low temperature combination (3 hours at 4±1°C) did not show any difference in terms of TPA parameters. In terms of appearance and general acceptability parameters, the scores of group B found higher than group A. In addition to that, odor, texture, spice, onion, and garlic taste variables of group B found more acceptable.

**Keywords:** Atlantic salmon, catering, meatballs, microbial transglutaminase (MTGase), color, sensorial analysis

### Atlantik Somonu (*Salmo salar* Linnaeus, 1758) Köftelerinin Fiziksel, Duyusal ve Besinsel Özellikleri Üzerine Mikrobiyal Transglutaminaz (MTGaz) Enziminin Etkisi

**Öz:** Bu çalışmada, ülkemizde ve özellikle dünya genelinde catering firmaları, oteller, restoranlar ve diğer hazır yiyecek işletmelerinin su ürünleri menülerinde yer alan Atlantik somonundan (*Salmo salar* Linnaeus, 1758), MTGaz ilavesi (%0.60) ile balık köftesi üretimi gerçekleştirilmiştir. Zamanın ve hijyenin oldukça önemli olduğu bu tür işletmeler için gıda güvenliğini riske atmamak adına kısa süre ve düşük sıcaklığın (4±1°C; 3 saat) kombine etkisinin MTGaz enzimi aktivasyonu için yeterliliğinin/uygulanabilirliğinin araştırılması amacıyla balık köftelerinde besinsel, fiziksel ve duyusal analizler gerçekleştirilmiştir. Üretim ve aktivasyon süreçlerinin ardından köfteler fırında 180 °C'de 20 dakika pişirilmiştir. MTGaz ilaveli grubun (B) ham protein ve nem içeriği kontrol (A) grubundan yüksek tespit edilirken (p<0.05), A grubunun enerji içeriği ise B grubuna oranla daha yüksek belirlenmiştir (p<0.05). A ve B gruplarının pH değerleri sırasıyla 7.40 ve 7.32 olarak tespit edilmiştir (p<0.05). Araştırmada, parlaklık (L\*) değerinin MTGaz kullanımı ile arttığı da ortaya koyulmuştur. TPA sonuçlarına göre; MTGaz ilavesi sonrasında pişirme öncesi düşük sıcaklıkta kısa süreli (4±1°C'de 3 saat) aktivasyon işleminin Atlantik somon köftesinin tekstüründe önemli değişikliklere neden olmadığı belirlenmiştir. Görünüş ve genel duyusal değerlendirme parametreleri bakımından B grubu daha yüksek puanlanmıştır. Ayrıca koku, tekstür, baharat, soğan ve sarımsak aroma değişkenlik parametreleri B grubunda daha beğenilir bulunmuştur.

**Anahtar Kelimeler:** Atlantik somonu, catering, köfte, mikrobiyal transglutaminaz (MTGaz), renk, duyusal analiz

#### 1. Introduction

Consumers around the world are supplied with aquaculture products through fishing and aquaculture. In the aquaculture sector, Atlantic salmon (*Salmo salar*) is the most widely farmed fish species. Approximately 90 million 500 thousand tons of fish were obtained in 2020,

of which 84 million tons were obtained from farms through controlled aquaculture. The farmed salmon farming industry, which started in the 1960s, has grown significantly over the past decades, and today about 70% of the salmon produced worldwide comes from aquaculture. In 2020, more than 2,600,000 tons of farmed

salmon were farmed worldwide, while only about 550,000 tons of natural salmon were caught (Statista, 2022a). Norway has the largest share in Atlantic salmon exports, while France, Singapore and Portugal are the leading importing countries (Statista, 2022b).

Türkiye is a country where the aquaculture sector is developing rapidly, and the export of aquaculture products has gained momentum in recent years. Trout, sea bass and sea bream are the most farmed and exported fish species in Türkiye. In addition, Türkiye imports a significant amount of Atlantic salmon from Norway (FAO, 2020). Between the years of 2014 and 2019, the average annual consumption of seafood per capita worldwide increased from 19.9 kilograms to 20.5 kilograms, but in 2020, seafood consumption fell to its lowest levels in recent years at 19.8 kg per capita (Statista, 2022c). In Türkiye, aquaculture consumption is quite below the world average. The main reasons for the low consumption in Türkiye are the popularity of seafood products and the prejudice of consumers against aquaculture fish. Additionally, the fact that fresh fish is more prominent in fish consumption and the lack of demand for processed seafood products.

While many product types such as canned, smoked, marinated, surimi, new generation packaged products, fish sausages, salami, burgers, etc. have found a place in foreign marketplaces, those products have not reached the deserved position in Turkish markets. The basis for increasing fish consumption in Türkiye can be realizable such as ensuring that children consume fish at least twice a week. Since the eating and drinking habits acquired in childhood will affect the whole life, the individual will continue this habit in later ages. It is essential that catering companies show the necessary sensitivity and plan their menus to include seafood 1 - 2 times a week so that children or young people who spend a large part of their day outside due to school and adults who are actively working can benefit from seafood at least at lunchtime. However, it is also known that seafood can become a risky food as it is very sensitive to spoilage due to its structure. It requires maximum attention, especially during preparation for mass consumption. In addition, there are quite high fluctuations in prices depending on the season. At this point, processed seafood products provide a solution to an important problem both in terms of industrialization and quality safety and cost fluctuations. For this reason, it is especially important to fillet large fish

such as salmon, remove the bones, form to minced meat and use them as meatball raw material in the sector.

One of the processed seafood products is fish meatballs or burgers. In its simplest definition, it is a product containing additives such as onion, breadcrumbs, salt, black pepper, garlic, red pepper, egg, etc. added to fish minced meat; in addition to the above-mentioned in the industrial sense; It may also contain potato starch, milk powder, enzyme, phosphate, carrageenan, preservative, and stabilizing agents, etc. (Can, 2012; Öksüztepe et al., 2010; Ulusoy et al., 2017). Transglutaminase enzyme is an enzyme used in meatball, burger, and pate production. Transglutaminase (TGase) initiates the formation of covalent bonds between glutamine and lysine residues in proteins. The addition of microbial transglutaminase (MTGase) can improve the thermal stability of meat proteins by imparting desirable properties to the reconstituted products during heating (Başaran et al., 2010). The best pH range for the enzymatic activity of MTGase is from 5 to 8. However, some enzymatic activity can be maintained at very low (~ 4) and very high (~ 9) pH levels (Motoki and Seguro, 1998). The optimum temperature for enzymatic activity is 50°C. However, it has also been reported that MTGase is active at 10°C and maintains some activity even at temperatures just above freezing (Motoki and Seguro, 1998). In the literature, there are studies on traditional and industrial meatballs produced from different fish species with different additives (Altan, 2020; Ehsani et al., 2020; Gökoğlu, 1994; Kaba et al., 2012; Keser and İzci, 2020; Kılınççeker, 2014; Kılınççeker and Karahan, 2019; Mattje et al., 2019; Özpolat and Çoban, 2012; Sugitha et al., 2019; Yanar and Fenercioğlu, 1999). In a previous study, it was determined that minced meat obtained from breeder-sized rainbow trout (*Oncorhynchus mykiss* Walbaum, 1792) can be preserved for up to 24 days in a quality manner by packing in modified atmosphere (MA) with 75%CO<sub>2</sub> + 25%N<sub>2</sub> combination (Kocatepe et al., 2016). Various studies have shown that sausages, meatballs, and burgers produced from fish with the addition of MTGase are highly appreciated and help to increase fish consumption. For example, Cavenaghi-Altémio et al. (2018) surveyed the "propensity to purchase" and "willingness to reconsume" of MTGase-added sausages produced from catfish. While 66% of the respondents answered, "I would definitely buy it", 23% answered "I would very likely buy it again". Another researcher (Altan, 2020) had a survey group that

were consisted of children between the ages of 8 and 15, who do not prefer to consume fish due to the presence of bones, skin, or its characteristic smell, or whose fish consumption rate is relatively low. After children were tried the trout burger patties produced with MTGase, they were reported that the “taste / enjoying of the product” and “wanting to consume the product again” criteria scores as more than 90%. According to the literature, it is obvious that various products such as fish sausages, burger patties, etc. with MTGase additives, which is a processed fish product, will contribute significantly to increase the fish consumption of the society, especially children.

In this study, it was aimed to determine the effect of activation of meatballs obtained from imported Atlantic salmon minced meat at  $4\pm 1^\circ\text{C}$  for 3 hours after the addition of MTGase on the nutritional, physical and sensory properties of meatballs.

## 2. Materials and Methods

### 2.1. Material

In this study, 5 Atlantic salmon (*Salmo salar* Linnaeus, 1758) (between 4 and 5 kg) were obtained from Samsun Fish Market. The fish were quickly transported to Sinop University, Faculty of Fisheries, Fisheries Processing and Quality Control Laboratory in Styrofoam boxes in ice.

### 2.2. Preparation of Fish Cakes

After the fish are brought to the laboratory, cleaned, filleted and the remaining skin on the fillets is removed. The bones were removed from the fillets and minced into minced meat by passing through a Mateka RR-K30D brand/model cutter (5000 r./min.) for 90 seconds. Approximately 2 kgs of minced fish were separated to create of two-groups. All additives were added to the minced meat and passed through the cutter for another 30 seconds to ensure complete mixing of the meatball mixture. Meatball mixture: 70% minced fish, 15% grated onion, 7.5% breadcrumbs, 5% egg, 1.30% salt, 0.85% garlic, 0.25% crushed red pepper and 0.20% black pepper. *Group A (control)*: Atlantic salmon meatballs without MTGase; *Group B*: Atlantic salmon meatballs with 0.60% MTGase enzyme addition. The groups were shaped as 5cm x 4cm x 1cm (length x width x thickness) and after resting at  $4\pm 1^\circ\text{C}$  for 3 hours, they were cooked in Ulubaş/UEO-TT/1400W brand/model oven at  $180^\circ\text{C}$  for 20 minutes. The study was carried out in 2 replicates and 3 parallels.

### 2.3. Analyses

Crude protein, crude lipid, crude ash, moisture, energy content, pH, water activity ( $a_w$ ), color analysis, texture profile analysis (TPA) and sensory analyses were performed. The analyses were carried out on cooked meatball samples.

#### 2.3.1 Proximate Composition Analysis

Dry matter, crude protein and crude ash analyses of the groups were carried out according to AOAC (1995) and crude lipid analysis was carried out in Gerhardt SE-416 brand/model fully automatic soxtherm oil extraction device according to AOAC (2000). Atwater method was used for energy calculation of fish balls (Falch et al., 2010). According to this method:

$$\text{Carbohydrate value} = 100 - (\text{Water} + \text{Lipid} + \text{Protein} + \text{Ash}) \quad (1)$$

$$\text{Energy (kcal)} = (\text{Lipid} \times 9) + (\text{Protein} \times 4) + (\text{Carbohydrate} \times 4) \quad (2)$$

#### 2.3.2. Physical analysis

The pH of the fish balls was measured with a digital pH meter of brand/model as Werkstätten 82362. 2 g of homogenized fish meat was homogenized in 20 ml of pure water (1:10 ratio) for 1 minute and the pH meter was immersed in this solution for 1 minute (Curran et al., 1981).

LabShift Novasina brand/model automatic water activity device was used to determine the water activity ( $a_w$ ). Measurements were carried out according to Horwitz et al. (1980) by placing approximately 5 g of sample in the measuring cup at  $25^\circ\text{C}$ .

Konica Minolta/CR-A 33a, brand/model colorimeter (CIE, 1976) was used for color measurement of fish balls. The samples were placed in transparent plastic petri plates with a diameter of 9 cm and the measurements were carried out in those petri plates. The \*a value indicates redness or greenness, \*b value indicates yellowness or blueness and \*L value indicates the degree of brightness between 0 (black) and 100 (white).

TPA (texture profile analysis) analysis was performed within the scope of mechanical analysis. Brookfield CT3 Texture Analyzer brand/model device was used. For TPA analysis, 1cm x 1cm x 1 cm (length x width x thickness) standard size samples were taken from cooked salmon meatballs cooled to room temperature. The device was set

to 60% deformation target, 0.05 N trigger sensitivity, 0.50 mm/sec test speed, 2 mm/sec pre-test speed, 20 points/sec data transfer rate and 50 kg cell load. A 12-mm-diameter cylindrical probe was used in the device. Among the TPA parameters; hardness (N), adhesiveness (mJ), resilience, cohesiveness, gumminess (N), chewiness (N.mm) and springiness (mm) values were measured (Anonymous, 2018).

All analyses were performed with 2 replicates 3 parallel (n=6).

### 2.3.3. Sensory analysis

As sensory analysis form of meatballs, the hedonic scale created by Pons et al. (2006) was modified according to the product and converted into a 5-point scoring system and odor, texture, appearance, spice, onion, garlic, general taste, and general rating parameters were asked to panelists. Cooked fish meatball evaluations were carried out by 20 panelists experienced in the field of aquaculture (with 2 replicates). Meatball samples were placed on 10 cm diameter white foam plates separately for each panelist and panelists were not informed about the meatball contents. In the scoring scale of cooked meatballs; 5 points were evaluated as "very good", 4 points as "good", 3 points as "medium", 2 points as "consumable" and 1 point as "very bad". A score of "3" on the raw trout scale and a score of "2" on the cooked trout scale was considered as the limit of consumability.

### 2.3.4. Statistical Analysis

Proximate composition, physical, color and TPA data were expressed as mean  $\pm$  standard error (se) and homogeneity of variances was tested at  $p < 0.05$  significance level. The *t-test* was used to compare the means of the analysis results of the groups. Statistical analyses were performed using MINITAB (Version 17.1) software. Within the scope of sensory analysis, paired sample t-test was used to compare the panelists' opinions on the differences between the groups using IBM SPSS (Version 21.4) software.

## 3. Results and Discussion

### 3.1. Proximate Composition of Fish Meatballs

Total crude protein, crude lipid, crude oil, crude ash, moisture, carbohydrate and energy contents of group A and B are given in Table 1.

While the crude protein and moisture content of the

MTGase-added group (B) was higher than that of group A (control) ( $p < 0.05$ ), the energy content of group A was higher than that of group B ( $p < 0.05$ ). Altan (2020) was reported that MTGase is preventing the moisture loss of trout burger patties due to structure enhancement and therefore, protein losses were seen at minimum levels. Although there was no statistical difference, the slightly higher crude lipid content of group A caused an increase in energy content. In terms of other nutritional analyses (crude lipid, crude ash, and carbohydrate), there was no difference between the groups ( $p > 0.05$ ).

Salmon is a fish species preferred by consumers due to its high lipid content. As it is known, lipids are one of the most important components affecting the taste of food/meal. In this study, no oil was added to the meatball batter considering the crude lipid content of salmon. Atlantic salmon has a higher lipid content compared to many natural fish species and aquaculture fish. In a study comparing the proximate composition of large rainbow trout burger patties with and without MTGase addition, it was reported that crude lipid content of patties decreased, crude protein content increased, and moisture content increased with 0.5% MTGase addition (Altan, 2020). In the same study, it was reported that the energy content of trout patties decreased with the addition of MTGase. In our study, the energy content of group A was found to be 209.33 kcal/100g and 194.18 kcal/100g in group B with MTGase addition and this value was higher than the energy content reported by Altan (2020). In the aforementioned study, 5% oil was added to the meatball mixture. Additives added to the meatball mixture affect the nutritional content of the final product.

Palmeira et al. (2014) reported that the crude protein content of trout meatballs increased with the addition of MTGase, similar with our study. In a study investigating the gel holding capacity of mackerel meat under high pressure with the addition of MTGase, it was reported that the protein solubility of the enzyme added group was lower (Gómez-Guillén et al., 2005). Similarly, Cardoso et al. (2011) reported that MTGase prevented protein loss in fish meat and the highest protein ratio was in the enzyme group. The use of MTGase reduces protein solubility in meatballs and enriches the final product in terms of protein content.

It is clear that the use of MTGase did not make a significant difference in the crude ash content of the groups. Similar with our study, Cardosa et al. (2011)

reported that the effect of MTGase supplementation on the crude ash value of minced sea bass fish was insignificant. MTGase enzyme is used in meat industry for different product development purposes. Uran et al. (2013) reported that the addition of 0.5% - 1% TGase enzyme to chicken breast meat caused an increase in protein and ash values and a decrease in lipid content, similar with our study.

Fish meat is a food with a very low carbohydrate content. Various researchers were reported the carbohydrate content of Atlantic salmon as 0.19g/100g (Şengör et al., 2013) and 1.72% (Kocatepe et al., 2022). In the production of Atlantic salmon meatballs, the additives added to the meatball batter increased the carbohydrate content of the final product up to 7.87 g/100g. This increase was seen lower in the group with MTGase addition ( $p>0.05$ ).

**Table 1.** Proximate composition and energy content of Atlantic salmon meatballs

**Çizelge 1.** Atlantik somonu köftelerinin besin kompozisyonu ve enerji içeriği

	<b>Group A (control)</b>	<b>Group B (0.60% MTGase)</b>
Crude protein (%)	14.80±0.07 <sup>B</sup>	15.19±0.03 <sup>A</sup>
Crude lipid (%)	13.18±0.58 <sup>A</sup>	11.77±0.22 <sup>A</sup>
Moisture (%)	61.71±0.12 <sup>B</sup>	63.82±0.04 <sup>A</sup>
Crude ash (%)	2.42±0.15 <sup>A</sup>	2.34±0.08 <sup>A</sup>
Carbohydrate (%)	7.87±0.57 <sup>A</sup>	6.86±0.33 <sup>A</sup>
Energy (kcal/100 g)	209.33±3.06 <sup>A</sup>	194.18±1.01 <sup>B</sup>

Mean ( $n=6$ ) ± std. error

A, B: (→) : The difference between groups with different letters is significant ( $p\leq 0.05$ ).

### 3.1. Physical Properties of Fish Meatballs

Physical analysis is one of the main factors affecting the preferability of food for the consumer. While some of the physical properties provide information about the composition of the food, some of them are important in the development of pre-consumption perception. Especially the color of the food directly addresses consumer perception. Table 2 shows the pH, aw, L\*, a\* and b\* color analysis results of Atlantic salmon meatballs.

The pH value of Atlantic salmon varies between 6.32 – 6.58 (Wang et al., 2003) and 6.13 – 6.20 (Duun & Rustad, 2008). The pH value of group A (without enzyme addition) was 7.40, while the pH content of the group with MTGase (B) was found as 7.32 ( $p<0.05$ ). Alkaline substances used in the meatball ingredients affected the pH content of the final product. It was reported by Motoki and Seguro (1998) that MTGase activity continues between pH 4 and 9. For MTGase activation, the

meatballs were rested at  $4\pm 1^\circ\text{C}$  for 3 hours and then cooked. Gaspar and Goes-Favoni (2015) reported that the addition of MTGase increased the pH level, and this was due to the high isoelectric point of MTGase itself. Karina and Setiadi (2020) reported that adding 1 – 1.5% MTGase to halibut fillets are directly increased the pH level sharply. In our study, the pH content of group A without enzyme addition was higher than group B. This difference between the studies is thought to be due to the enzyme ratio and the content of meatball ingredients used in our study.

**Table 2.** Physical properties of Atlantic salmon meatballs  
**Çizelge 2.** Atlantik somonu köftelerinin fiziksel özellikleri

	<b>Group A (control)</b>	<b>Group B (0.60% MTGase)</b>
pH	7.40±0.01 <sup>A</sup>	7.32±0.00 <sup>B</sup>
A <sub>w</sub>	0.95±0.00 <sup>B</sup>	0.96±0.00 <sup>A</sup>
<i>Color analysis</i>		
L	74.34±0.94 <sup>B</sup>	76.08±0.12 <sup>A</sup>
a*	6.28±0.16 <sup>A</sup>	5.37±0.22 <sup>B</sup>
b*	22.78±0.07 <sup>A</sup>	22.01±0.25 <sup>B</sup>

Mean ( $n=6$ ) ± std. error

A, B: (→) : The difference between groups with different letters is significant ( $p\leq 0.05$ ).

Water activity ( $a_w$ ), which is a measure of the amount of moisture in foods, is directly related to the moisture content of the food. The  $a_w$  content of fish meat varies between 0.97 – 0.99. With the addition of enzyme, it was also aimed to get a product with a higher water content. As mentioned before, the moisture content of group B containing enzyme was higher than group A. Similarly, water activity was also higher than the group without enzyme ( $p<0.05$ ). It is obvious that some water is removed from the meatball content with the effect of cooking, but despite this, the  $a_w$  content of the meatballs is 0.95 and above.

The color of food is a feature that gives a lot of information to the consumer about the content and general characteristics of the product and reflects the deterioration parameters of the food. Since color perception can vary from person to person, color results obtained with laboratory devices are more reliable. L\* is expressed as brightness value. As seen in Table 2, the L\* value of the enzyme-added group was found higher ( $p<0.05$ ) than control group. Similar with our study, it has been reported by different researchers that the brightness value increases with the use of MTGase due to the catalyzing the cross-linking reaction of myosin leading to the formation of

protein intra- and inter- molecular covalent bonds, and it is resulting with formation of opaque gels (Karayannakidis et al., 2008; Ramirez et al., 2000). +a\* value expresses reddish color while +b\* color expresses yellow color. Both +a\* and +b\* values of group B were found lower ( $p < 0.05$ ). Cavenaghi-Altemio et al. (2018), were examined the color quality effect of MTGase addition at different ratios on catfish sausage and they stated that +a\* and +b\* values of low enzyme concentrations were higher, while these values decreased with enzyme increase.

The results of texture profile analysis (TPA) of Atlantic salmon meatballs are shown in Table 3. Brookfield Laboratories, which specializes in texture and has carried out very important studies in this field, defines TPA parameters in its manual as follows: Hardness is the deformation response to the analysis probe; adhesiveness is the tendency of the food to adhere to the teeth during the first bite; resilience is the effort to return to its initial state after the food is compressed in the first cycle; cohesiveness is the resistance of the food to a second deformation; gumminess is the force required to chew solid food to make it ready for swallowing; and springiness is the flexibility of chewing at the first moment in the mouth (Brookfield Inc., 2018). TPA analysis is a kind of laboratory simulation of the chewing force applied by the jaw muscles and teeth when the food is taken into the mouth. For this reason, properties of foods such as hardness, resilience, cohesiveness, etc. are measured by texture analysis and give clear results.

When the TPA findings of the groups were examined, no difference was detected in any factor ( $p > 0.05$ ) except for the resilience ( $p < 0.05$ ). Although no statistical difference was detected, the springiness values of group B were found increased. Similarly with our research, Altan (2020) reported that the springiness of cooked trout burger patties produced with MTGase was found higher in the groups with 0.5% and 1% enzyme addition compared to the control group. According to the TPA results of the study, it was determined that short-term (3 hours at  $4 \pm 1^\circ\text{C}$ ) resting at low temperature before cooking after the addition of MTGase did not cause significant changes in the texture properties of Atlantic salmon meatballs. This may be related to the fact that 3 hours of resting at  $4 \pm 1^\circ\text{C}$  was not sufficient for the activation of the enzyme. Tokay

et al. (2022) reported that short setting time (4 hours) was not efficient for the enzyme to activate and restructure fish meat. It can be suggested that the enzyme activation time and temperature should be increased for future studies such as meatballs/burgers/pate.

**Table 3.** Texture profile analysis (TPA) characteristics of Atlantic salmon meatballs

**Çizelge 3.** Atlantik somonu köftelerinin tekstür profil analizi (TPA) bulguları

	Group A (control)	Group B (0.60% MTGase)
Hardness (N)	20.15±1.62 <sup>A</sup>	20.13±0.32 <sup>A</sup>
Adhesiveness(mJ)	0.55±0.05 <sup>A</sup>	0.55±0.55 <sup>A</sup>
Resilience	0.07±0.00 <sup>A</sup>	0.05±0.01 <sup>B</sup>
Cohesiveness	0.26±0.00 <sup>A</sup>	0.26±0.02 <sup>A</sup>
Gumminess (N)	5.30±0.38 <sup>A</sup>	5.24±0.56 <sup>A</sup>
Chewiness (N.mm)	46.9±11.9 <sup>A</sup>	46.85±13.25 <sup>A</sup>
Springiness (mm)	9.98±0.39 <sup>A</sup>	10.24±0.07 <sup>A</sup>

Mean (n=6) ± std. error

A, B: (→) : The difference between groups with different letters is significant ( $p \leq 0.05$ ).

### 3.2. Sensorial Characteristics of Fish Meatballs

In the sensorial analysis results (Table 4), which was conducted to determine whether the panelists' ratings of the groups on the 5-point hedonic scale differed or not, it was observed that the differences between the mean ratings were observed between the variables of General Taste ( $p = 0.000 < 0.05$ ); Appearance ( $p = 0.021 < 0.05$ ) and General acceptability ( $p = 0.000 < 0.05$ ).

**Table 4.** Paired sample t-tests statistics of sensory analyses

**Çizelge 4.** Duyusal analiz verilerinin eşleştirilmiş örneklem t-testi sonuçları

		Average Panelist	Std. Deviation	Mean of Std. Error
Pair 1	Odor A	4,25	20	0,71635
	Odor B	4,35	20	0,74516
Pair 2	Texture A	4,20	20	0,52315
	Texture B	4,40	20	0,68056
Pair 3	Spice taste A	3,85	20	0,74516
	Spice taste B	4,25	20	0,63867
Pair 4	Onion taste A	3,65	20	0,81273
	Onion taste B	3,95	20	0,99868
Pair 5	Garlic taste A	3,75	20	1,01955
	Garlic taste B	4,10	20	0,85224
Pair 6	General taste A	3,80	20	0,76777
	General taste B	4,58	20	0,67424
Pair 7	Appearance A	3,95	20	0,68633
	Appearance B	4,45	20	0,68633
Pair 8	General acceptability A	3,93	20	0,73045
	General acceptability B	4,70	20	0,47016

**Table 5.** Paired sample t-tests of sensory analyses, paired differences**Çizelge 5.** Duyusal analizlerin eşleştirilmiş örneklem t-testi farklılıkları

		Average	Std. Deviation	Std. Error	95% Confidence interval			t	df	Sig. (2-tailed)
					Average	Low	High			
Pair 1	Odor A - Odor B	-,10000	,55251	,12354	-,35858	,15858	-,809	19	,428	
Pair 2	Texture A - Texture B	-,20000	,76777	,17168	-,55933	,15933	-1,165	19	,258	
Pair 3	Spice taste A – Spice taste B	-,40000	,99472	,22243	-,86554	,06554	-1,798	19	,088	
Pair 4	Onion taste A – Onion taste B	-,30000	1,08094	,24170	-,80589	,20589	-1,241	19	,230	
Pair 5	Garlic taste A – Garlic taste B	-,35000	1,18210	,26433	-,90324	,20324	-1,324	19	,201	
Pair 6	Gen. taste A - Gen. taste B	-,77500	,73404	,16414	-1,11854	-,43146	-4,722	19	,000	
Pair 7	Appearance A - Appearance B	-,50000	,88852	,19868	-,91584	-,08416	-2,517	19	,021	
Pair 8	Gen. accept. A - Gen. accept. B	-,77500	,76906	,17197	-1,13493	-,41507	-4,507	19	,000	

There was no difference between the other variables ( $p>0.05$ ). When the mean scores of the variables in which differences were observed are examined, it is possible to say that the mean scores of Group B in the general taste parameter are higher than Group A ( $4.57>3.80$ ) and likewise, Group B is evaluated by the panelists with higher scores in the parameters of Appearance ( $4.45>3.95$ ) and General acceptability ( $4.70>3.9250$ ) compared to Group A.

On the other hand, when the average scores given by the panelists to the products were examined in general terms, it was found that Group B had higher average scores than Group A in terms of odor, texture, spice, onion, and garlic taste variables (Table 4 and 5). According to these results, Group B was found to be more acceptable by the panelists than Group A.

#### 4. Conclusion

When the results of the research are evaluated, it can be said that although a relatively low amount (0.60%) of MTGase was used, it significantly protected the product quality compared to the control (A) group by reducing crude protein and moisture loss. In the control (A) group, the total energy amount increased due to the slightly higher crude lipid content. When the pH and aw differences between the groups are examined, even though statistical differences were observed in both analyses, the differences are not of any practical significance. When the color changes between the groups were examined, it was found that the use of enzyme increased the brightness ( $L^*$ ) while decreasing the redness ( $a^*$ ) and yellowness ( $b^*$ ) values. In terms of texture profile analysis (TPA) findings, the groups showed very similar results to each other. The main reason for this similarity is clearly seen to be the reverse synergistic effect of low temperature and limited time (3 h at  $4\pm 1^\circ\text{C}$ ) on enzyme activation. It is possible to

say that group B containing MTGase had higher scores than the control (A) group in terms of appearance, general taste, and general acceptability parameters, as well as higher scores than the control (A) group in terms of odor, texture, spice, onion, and garlic variables. According to these findings, group B containing MTGase achieved a higher level of appreciation by the panelists than the control (A) group.

To evaluate the research as a whole, although the MTGase enzyme showed low activity due to low temperature/time application and could not be effective on textural properties, it is still managed to provide preservation in an important component of the nutritional composition such as protein, and to preserve the color properties to a great extent and to gather all the appreciation in terms of all other sensory criteria. Considering that the biggest goal of the sector is to gain customer appreciation and that consumer preferences are based on product appearance, it can be said that Group B was very successful in providing the most important criteria for consumers (color, taste, general appearance, etc.). In future research, it may be recommended to develop new products with MTGase additives by increasing the activation temperature and duration slightly more without risking food safety.

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