

Definition of textural deterioration in squid samples: Three different tools supported by microbial, visual and physico-chemical analysis

Kalamar örneklerinde tekstürel bozulmanın tespiti: üç farklı aracın mikrobiyolojik, görsel ve fiziko-kimyasal analizlerle desteklenmesi

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Abstract: The aim of the present study was to reveal the textural profile changes like hardness, cohesiveness, springiness, and adhesion supported by total mesophilic aerobic bacteria count (TMABc), pH, and some visual sensory characteristics in squid samples stored at 4 °C. Three different Brookfield Texture Analyzer tools, named TA7, TA9, and TA18 were used to observe the textural changes more clearly. The difference of TMABc between the storage days reached from 4.32 log CFU/g to 6.32 log CFU/g. The hardness value of the squid samples, detected by TA18 and TA9 tools, increased while the hardness value obtained from TA7 was higher. The highest change in cohesiveness value in the squid samples was defined by the TA9 tool as ~63%. Once springiness values detected by TA9 were sharply decreased from 4.9 mm to 2.1 mm, those of TA7 and TA18 were slowly decreased. The most increase in adhesion value (0.08 mJ to 0.21 mJ) was obtained in the TA7 tool. Depending on the textural quality changes, the pH value was increased, and as visual and sensory, dark, or yellow spots were observed. The present study results revealed that especially the TA18 tool could be effectively used to determine the quality changes of the squid samples.

Keywords: Texture, hardness, cohesiveness, springiness, adhesion, squid

Öz: Bu çalışmanın amacı, 4 °C'de depolanan kalamar örneklerinde toplam mezofilik aerobik bakteri sayısı (TMABc), pH ve bazı görsel duyu özelliklerinin desteklediği sertlik, kohezyon, yayılma ve yapışma gibi dokusal profil değişikliklerini ortaya çıkarmaktır. Tekstürel değişiklikleri daha net gözlemlemek için Brookfield Tekstür Analiz cihazının TA7, TA9 ve TA18 kodlu üç farklı aracı kullanılmıştır. Depolama günleri arasındaki TMABc 4,32 log KOB/g'dan 6,32 log KOB/g'a ulaşmıştır. TA18 ve TA9 araçlarıyla tespit edilen kalamar numunelerinin sertlik değeri artarken TA7'den elde edilen sertlik değeri daha yüksek olmuştur. Kalamar örneklerinde kohezif yapışkanlık değerindeki en yüksek değişiklik TA9 aracı ile ~%63 olarak tanımlanmıştır. TA9 tarafından tespit edilen elastikiyet değerleri, 4,9 mm'den 2,1 mm'ye keskin bir şekilde düşerken, TA7 ve TA18'in değerleri daha yumuşak bir azalış göstermiştir. Adezif yapışkanlık değerinde en fazla artış (0,08 mJ'den 0,21 mJ'ye) TA7 aracıyla elde edilmiştir. Tekstürel değişimlerle birlikte, pH değeri artmış, üründe bölgesel karama veya sarı lekeler gözlemlenmiştir. Bu çalışmanın sonuçları, kalamar örneklerinin kalite değişikliklerini belirlemede, özellikle TA18 aracının mikrobiyolojik ve fiziko-kimyasal analizlerle, daha ileri kalamar doku çalışmalarına rehberlik edebileceğini ortaya koymuştur.

Anahtar kelimeler: Doku, sertlik, kohezif yapışkanlık, elastikiyet, adezif yapışkanlık, kalamar

INTRODUCTION

Seafood is a food group with high protein value, rich in omega-3 fatty acids such as EPA, DHA and unsaturated fatty acids (Watters et al., 2012). Numerous studies have shown that people who consume seafood have a lower risk of developing diabetes and cardiovascular disease (Ceylan, 2014). Also, fish meat is known as the most consumed food material among seafood. However, there has been an increase in the interest in shellfish in recent years as well. Especially the consumption of shrimp and squid plays an important role among them. Since seafood is highly perishable, the rapid increase in some quality changes such as microbiological, sensory, chemical, and textural deterioration can be more easily observed (Cheng et al., 2014; Rodrigues et al., 2017; Yu et al., 2017). Like in many food materials, the growth in total mesophilic bacteria is used as an indicator to determine the shelf life of seafood (Armani et al., 2016; Fazlara et al., 2014).

Especially microbial growth in seafood and sensory properties can give an idea to evaluate the products for the consumers. Moreover, the changes in sensory characteristics like odor could be linked with microbiological spoilage in seafood (Shalini et al., 2015; Sutikno et al., 2019). In this respect, when the squid samples are thought to be among the highly perishable seafood, particularly the rapid changes in the color of the squid samples reveal the quality changes of the products during the cold storage period. For example, occurring pink spots on the surface of the squid samples could be easily detected (Sungsri-in et al., 2011). Besides the parameters which can be rapidly evaluated by the consumers, the changes in pH can have an important role in the squid samples (Márquez-Ríos et al., 2007). The squid samples are treated with different processing applications and the pH or textural properties could be affected before consumption. Also, textural applications in food science are named as one

of the practical methods that provide lots of advantages such as cost-effectiveness and giving speedy results with spending less time. Textural properties can define the preferences of the consumers as well. Analytical tools applied to evaluate fish freshness, such as chemical and microbiological analyzes, are time-consuming and require various chemicals (Rodrigues et al., 2017). Texture analyzes are quick, expertise-free, and non-destructive analyzes. With these analyzes, accurate assessment of fish quality, shortening the analysis time, and preventing sample loss can be provided. In addition, more sensitive results are obtained from subjective measurements based on human senses. Despite these advantages, there are very few studies using texture analysis as an indicator of fish freshness quality (Ceylan and Meral, 2018; Ceylan, et al., 2020a). This case is acceptable for the squid samples as well.

Besides the above-mentioned properties of squids, squid samples are widely exported globally. Therefore, squid has economical importance for the public as well. In this sense, initial quality, or the observation of the quality changes of squid samples stored at usually -18 °C and then thawing at 4 °C are very significant for the economic model of the countries and for the food safety. Thusly, the main aim of the present study was to reveal the microbial quality changes of squid samples stored at -18 °C and then thawed at 4 °C for getting ready consumption. Also, it is widely known that the textural properties of squid are also important, so the second target of the study was to investigate the different textural properties of the same samples by using three different texture tools. The final goal of the study was to support the textural changes with the pH, sensory, photographic images, and microbiological changes for the consumers and food industry.

MATERIAL AND METHODS

Materials

Frozen imported squid (*Uroteuthis duvauceli*) samples were obtained from the wholesale market at -18 °C, transported to food processing department in an icebox, thawed, and stored at 4 °C (Figure 1). Stored samples were analyzed for four days at room temperature.



Figure 1. Fresh squid sample

Methods

Total mesophilic aerobic bacteria count: TMABc in squid samples were determined. For each sample, 10 g squid was homogenized for 150 s in a stomacher (HG 400 V, Mayo International srl, Novate Milanese, Italy) with 90 mL peptone water (0.1%). Serial dilutions were prepared from 10^{-1} to 10^{-6} for each sample.

1 ml from the diluted sample was placed plate count agar (Merck-VM888763 930) by cast plate method and incubated (WiseCube® Wisd Digital Incubator WIG-50, DAIHAN Scientific Co., Ltd, South Korea) at 35 °C for 48 h to estimate TMABc in squid samples stored at 4 °C for 4 days (Maturin and Peeler, 2001).

pH: Before the pH measurements, all samples were homogenized using WiseTis HG-15D Digital Homogenizer (DAIHAN Scientific Co., Ltd, South Korea) separately. The homogenized squid samples were stirred by using distilled water (1:10). pH values were measured with a pH meter (HANNA HI9124, Hanna Instruments, Romania). The pH analysis was repeated for the samples three times.

Photographic and quality evaluation: A well-ventilated and lighted sensory analysis room were presented to the judges for better observation of the sensory changes. In this study, comments related to the quality evaluation are presented. Photographic images were obtained by using a cell phone having 3024x4032 pixels and 72 dpi.

Texture Analysis: For texture analysis, Brookfield CT3-1000 (Brookfield Engineering Laboratories, Middleboro, USA), Accessories Rotary Base Table (TA-RT-KIT), Ball: (TA18 12.7 mm diameter, stainless steel, 30 g), Needle: (TA9, 1.0 mm diameter, 43 mm length, 10° Maximum taper) and Knife-edge: (TA7, 60 mm wide, clear acrylic, 8 g) were used in the present study (Figure 2). Test parameters for Ball, Needle and Knife-edge measurements were determined as Trigger Value (TV) 0.03 N, Deformation (D) 2 mm, TV 0.03 N, D 5 mm, and TV 0.06 N, D 4 mm, respectively. In this respect, the changes in hardness (N), cohesiveness, springiness (mm), and adhesion (mJ) properties of the samples were revealed during the experimental period. In order to reveal the textural changes in the squid samples, a method by Ceylan et al. (2020a) was modified and used in the present study. The analysis was repeated for the samples in 10 mm depth three times at room temperature.



Figure 2. Ball (TA18), needle (TA9), and knife-edge (TA7) probes left to right

Statistical evaluation

For statistical analysis, all measurements were repeated with three replications. Obtained data were subjected to analysis of variance (ANOVA) to evaluate the textural changes, pH, and microbial growth every analysis day. GraphPad Prism Software Version 5.00 (California Corporation, CA) was performed to reveal significant differences between the analysis days, and comparisons of all differences among them were evaluated by Tukey's Multiple Range Test ($p < 0.05$).

RESULTS AND DISCUSSION

Textural changes

Hardness: Textural changes revealed by hardness, cohesiveness, adhesion, and springiness properties of imported squid samples are given in Table 1, 2, and 3. Hardness value is defined as the strength of food to resist the applied force according to Ertaş and Doğruer (2010). The hardness results obtained TA18 probe (Table 1) revealed that depending on the increase in storage period, hardness values of the samples were increased from 0.5 N to 3.39 N ($p < 0.05$).

Table 1. Texture measurements with ball (TA18) probe depending on the storage time

Analysis Days	0. Day	3. Day	4. Day
Hardness (N)	0.5 ± 0.17 ^c	1.64 ± 0.09 ^b	3.39 ± 0.26 ^a
Cohesiveness	0.57 ± 0.03 ^b	0.63 ± 0 ^a	0.795 ± 0.2 ^a
Springiness (mm)	1.6 ± 0.14 ^a	1.35 ± 0.07 ^b	1.35 ± 0.21 ^b
Adhesion (mJ)	0.06 ± 0.05 ^c	0.1 ± 0.01 ^b	0.17 ± 0.006 ^a

a, b, c letters define the statistical significant differences between storage days ($p < 0.05$)

Also, an increase in hardness value obtained from the TA9 probe (Table 2) was observed ($p < 0.05$). But, the ratio in an increase of TA18 and TA9 was different from each other. In another word, a rapid and sharp increase was observed using the TA18 probe.

Table 2. Texture measurements with needle (TA9) probe depending on the storage time.

Analysis Days	0. Day	3. Day	4. Day
Hardness (N)	0.84 ± 0.04 ^c	0.9 ± 0.07 ^b	1.06 ± 0.21 ^a
Cohesiveness	0.89 ± 0.11 ^a	0.33 ± 0.18 ^b	0.51 ± 0.32 ^b
Springiness (mm)	4.9 ± 0.00 ^a	2.1 ± 0.14 ^b	2.6 ± 0.7 ^b
Adhesion (mJ)	0.15 ± 0.025 ^a	0.20 ± 0.025 ^a	0.18 ± 0.035 ^a

a, b, c letters define the statistical significant differences between storage days ($p < 0.05$)

On the other hand, according to the results of TA7 (Table 3), the increase of the storage period was associated with the decline in hardness value (6.13 N to 1.28 N).

Table 3. Texture measurements with knife-edge (TA7) probe depending on the storage time

Analysis Days	0. Day	3. Day	4. Day
Hardness (N)	6.13 ± 0.93 ^a	2.34 ± 0.99 ^b	1.28 ± 0.16 ^c
Cohesiveness	0.57 ± 0.09 ^a	0.6 ± 0.02 ^a	0.59 ± 0.04 ^a
Springiness (mm)	1.80 ± 0.14 ^a	1.50 ± 0.00 ^c	1.65 ± 0.07 ^b
Adhesion (mJ)	0.08 ± 0.03 ^c	0.10 ± 0.02 ^b	0.21 ± 0.015 ^a

a, b, c letters define the statistical significant differences between storage days ($p < 0.05$)

In this sense, Cheng et al. (2014) noted that softness would be evaluated as a loss for texture in seafood. Also, some studies reported that depending on the storage, hardness was decreased (Alasalvar et al., 2001; Jain et al., 2007). These declines in hardness are widely associated with chemical and enzymatic reactions as stated by Chéret et al. (2005). In this respect, the probe type in the present study played a key role in order to detect the changes or deterioration in imported squid samples. Furthermore, the hardness values of the samples were detected to be unstable during the experimental period.

Cohesiveness: The cohesiveness value of the samples was also investigated using three different probes. In this respect, Mousavi et al. (2019) stated that cohesiveness is determined to be the forces of inner bond links that maintain the product as perfect, and it could be expressed as the force content, which could cause deform a material. Also, this value could be associated with consumers' preferences. According to the results of the present study, the initial value of the samples, which was detected by the TA18 probe was determined to be 0.57, but at the end of the storage period, that was found as 0.795 ($p < 0.05$). Also, similarly, TA9 probe results showed that depending on the increase in the storage period, cohesiveness was decreased from 0.89 to 0.33 ($p < 0.05$). On the other hand, TA7 was not found to be effective to determine the quality changes based on cohesiveness.

Springiness: In addition to cohesiveness and hardness, springiness value in the imported squid samples was observed. All probe types revealed that springiness value was declined. Furthermore, this decline was carried out for ball probe (TA18) as 15.6% (1.6 to 1.35 mm). For needle probe (TA9), this decline was observed in the range of 57.14% (4.90 to 2.1 mm) and for knife-edge probe (TA7), it was 8.33% (1.80 to 1.65 mm). Sutikno et al. (2019) reported that the springiness value of raw squid samples was 1.14 mm. According to Narasimha Murthy et al. (2018), icing methods such as chilling onboard or slurry ice could play a key role to determine the springiness value. Also, different types of

probes could be effectively used to define the quality changes in the squid samples.

Adhesion: It was observed that adhesion values of the imported squid samples were increased. These increases revealed by TA18 and TA17 probes were defined to be rapid as compared to the results of the TA9 probe. Also, there were statistical differences between the storage periods ($p < 0.05$). Moreover, in TA7, the initial value was increased from 0.08 mJ to 0.21 mJ at the end of the experimental period. Dunnewind et al. (2004) noted that there might be a correlation between adhesion value and sensory changes in some foods. So, investigation of AD values could be much more important for the squid consumers.

Total mesophilic aerobic bacteria growth

TMABc of the imported squid samples during the experimental period are given in Table 4. As known, TMAB growth in food samples plays a key role to define the quality (Ceylan, et al., 2020b). In this respect, besides the definition of textural quality by using three different texture probes of the imported squid, the relationship between textural deterioration and microbial spoilage is gaining today much more important in food science (Ceylan, et al., 2020a). In this study, the initial TMAB load of the imported squid samples was found as 4.39 log CFU/g. In this period, higher textural results were obtained as compared with the other days of the storage period. By the increase of the time, TMABc began to increase and on the 3rd day of the cold storage it reached 5.28 log CFU/g, and then in the last period, TMAB load of the imported squid samples had 6.30 log CFU/g. Bacterial spoilage in squid can cause other chemical amines such as biogenic, which can be hazardous for the consumers (Kim et al., 2009). Already, while the TMABc of the imported squid samples was increasing, the adhesion value was also increasing. However, the springiness value obtained from three different probes was significantly decreased. Furthermore, cohesiveness and some hardness values were depended on the probe type. For example, in hardness, according to the TA18 probe results, the value was rapidly increased with the increase of TMABc. But, when TA7 values were declined also, TMABc was increasing. Therefore, the study especially revealed that the used probe-type should be illustrated with the microbiological growth in the squid samples.

pH

The changes in pH values of the imported squid are presented in Table 4. The pH value could be evaluated as

one of the most effective indicators of meat quality as described by Gokoglu et al. (2017). Besides revealing the potential relationship between the TMAB growth and textural changes in the squid samples, observation in the pH changes could be evaluated as an important parameter. In this regard, at the beginning of the storage period, the pH value was detected to be 7.88. When the TMABc reached the highest value, the pH value was also reached 8.16. It could be observed that the pH value might decrease after death because of converting from glycogen to lactic acid in meat and then an increase could be widely detected with the increased time (Ceylan et al., 2018). Also, by this study, the increase in the pH value of the squid could be clearly associated with the increase in adhesion and the decrease in springiness.

Table 4. Quality parameters and changes for squid samples in regards to storage time.

Analysis Days	0. Day	3. Day	4. Day
pH	7.88 ± 0.01 ^b	7.76 ± 0.03 ^c	8.16 ± 0.03 ^a
TMAB (log CFU/g)	4.39 ± 0.09 ^c	5.28 ± 0.08 ^b	6.30 ± 0.01 ^a
Observations	Hard, bright structure	Mild smell, water release, color fading, formation of spots,	Rubberization, stickness, drying, growth of yellow spots,

a, b, c letters define the statistical significant differences between storage days ($p < 0.05$)

Photographic and quality evaluation

Figure 3 reflects the photographic changes in the squid samples depending on storage time. As can be seen from the figure, there was a smooth and clear meat surface at the beginning of the study. In another word, there were no yellowish, pink, or reddish structures on the surface of the squid meat. Particularly, depending on the loss in the textural properties, different colors and spots were defined on the surface of the squid samples. These images had a good relationship with quality evaluation as well. Because off-odor, dried and the changes in hardness, which may occur in loss of water were clearly seen. As know that different colorimetric measurements such as image analysis are used to define the quality of the meat products (Ceylan et al., 2018; Ünal Şengör et al., 2019). But, instead of costly methods, simple applications like textural measurement are recently preferred to determine the quality of food products. Moreover, this kind of study may play a guiding role to be built data-base systems for textural properties such as hardness, cohesiveness, springiness.



Figure 3. Visual changes of squid samples on day 0, 3, and 4

CONCLUSION

Different texture tools (TA7, TA9, and TA18) were effectively used to reveal the textural changes. TMABc of the squid samples were increased with time, at the same analysis periods, the textural changes were also observed by the three tools in the same period. Particularly, the clear changes in the hardness value were more successfully revealed by TA18 and TA9 probes.

During the analysis period, the changes of the cohesiveness value in the squid samples were observed as

~63% by the TA9 tool. Sharply decrease in springiness value was also obtained with the TA9 probe. On the other hand, the TA7 tool showed that adhesion value could increase by the increase of the storage period. In terms of the observation of the textural changes, the use of three different textural tools provided clear, understanding, and comprehensive opportunities. These changes were also observed by the other quality changes such as sensory, visual, and pH. The present study results suggested that the use of the TA18 probe could be more efficient as compared to the others.

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