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Research Article

The Effect of Different Storage Conditions on Textural, Microbiological and Color Properties of Phyllo (Yufka)

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Abstract

The effects of different storage conditions on textural, microbiological and color properties of phyllo were investigated. Phyllo samples were stored at room temperature (20ºC) for 3, 5 and 7 days, in refrigerator (4ºC) for 7, 14 and 21 days, and in deep freezer (-18ºC) for 20, 40 and 60 days. It was determined that phyllo samples stored in the deep freezer exhibited higher L* color values (82.33-82.97), followed by samples stored in the refrigerator (78.53-81.75) and samples stored at room temperature (75.74-77.75), respectively. A statistically similar and lower total color change (ΔE) occurred in the phyllo samples stored at room temperature and in the refrigerator compared to the samples stored in the deep freezer. The samples stored in the refrigerator exhibited higher hardness, chewiness and gumminess values, while the samples stored in the deep freezer exhibited lower values. Most of the samples produced were similar to the control sample in terms of springiness property. In terms of relaxation time, the samples stored at room temperature and in the deep freezer exhibited statistically similar and lower values than the samples stored in the refrigerator. Additionally, the samples stored in the deep freezer had higher breaking force and breaking length values. The total mesophilic aerobic bacteria count of the samples varied between 3.38-12.30 log cfu/g, and mold and yeast count varied between <2.00-6.87 log cfu/g.

Keywords: Phyllo (yufka), storage, textural properties, microbiological properties, color

Farklı Depolama Koşullarının Yufkanın Dokusal, Mikrobiyolojik ve Renk Özellikleri Üzerine Etkisi

Öz

Farklı depolama koşullarının yufkanın dokusal, mikrobiyolojik ve renk özellikleri üzerine etkisi araştırılmıştır. Yufka örnekleri oda sıcaklığında (20ºC) 3, 5 ve 7 gün, buzdolabında (4ºC) 7, 14 ve 21 gün ve derin dondurucuda (-18ºC) 20, 40 ve 60 gün boyunca depolanmıştır. Derin dondurucuda depolanan yufka örneklerinin daha yüksek L* renk değerlerine (82.33-82.97) sahip olduğu ve bunu sırasıyla buzdolabı (78.53-81.75) ve oda sıcaklığında depolanan örneklerin (75.74-77.75) izlediği belirlenmiştir. Oda sıcaklığı ve buzdolabında depolanan yufka örneklerinde, derin dondurucuda depolanan örneklere kıyasla istatistiksel olarak benzer ve daha düşük seviyede toplam renk değişimi (ΔE) meydana gelmiştir. Buzdolabında depolanan örnekler daha yüksek, derin dondurucuda depolanan örnekler daha düşük sertlik, çiğnenebilirlik ve sakızımsılık değerleri sergilemiştir. Üretilen örneklerin çoğu elastikiyet özelliği bakımından kontrol örneğine benzerdir. Gevşeme süresi bakımından oda sıcaklığında ve derin dondurucuda depolanan örnekler buzdolabında depolanan örneklere göre istatistiksel olarak benzer ve daha düşük değerler göstermiştir. Ayrıca derin dondurucuda depolanan örnekler daha yüksek kopma kuvveti ve kopma uzunluğu değerlerine sahiptir. Örneklerin toplam mezofilik aerobik bakteri sayısı 3.38-12.30 log kob/g ve küf ve maya sayısı ise <2.00-6.87 log kob/g arasında değişmiştir.

Anahtar Kelimeler: Yufka, depolama, dokusal özellikler, mikrobiyolojik özellikler, renk

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1. Introduction

Cereals and cereal-based products have an important role in human nutrition as they provide a significant portion of the daily energy needed [1]. Phyllo, also called yufka or fyllo, has a special position among cereal-based products due to its being a ready-to-use, practical [2] and easily accessible product and creating a feeling of fullness. It is a semi-finished product obtained by rolling out and partially baking the dough prepared using baklava and pastry type of wheat flour, drinking water, edible salt and additives when necessary [3]. Phyllo is a single-layered, cream-colored and 1–2 mm thick product [4] and can be produced in different shapes and sizes according to need [2].

Phyllo attracts attention especially due to its practical use [5] and is used as an intermediate product in the preparation of many pastries such as börek, flatbread [2], baklava, and pies like kasseropita and spanakopita [6], as well as desserts, samosas, and other delicious dishes [7]. It offers the opportunity to produce various food products in a short time and is widely preferred by consumers in modern living conditions, especially by people who have a busy work life and limited time to spare for food preparation and consumption [5]. For this reason, the demand for phyllo, which already has a wide consumption geography spanning the Middle East, Balkans, Caucasus and Turkey, is constantly increasing [8]. It is also stated that phyllo has become one of the most popular foods in many US, Asian and European countries due to its ease of cooking and taste [7, 9].

The most important quality characteristics of phyllo are surface color, proper baking degree and uniform diameter and thickness [10]. Additionally, textural properties are known to be extremely important for phyllo, especially in terms of the quality of the final product to be prepared using phyllo. It is expressed that phyllo must have perfect springiness and flexibility. It must also have excellent extensibility, all of which are desirable to provide the ability to fold and roll during processing [2, 4, 10]. In addition, phyllo should be neither be sticky nor so weak that it tears easily. If phyllo does not meet the appropriate textural properties, the final products cannot be properly produced, or even if they are produced, significant quality problems may arise in the final product, leading to substantial economic losses.

Phyllo generally has a short shelf life due to its neutral pH (6-7) and comparatively high water activity (0.6-0.8 aw), which make it a suitable substrate for aerobic microorganisms. This requires keeping the phyllo under appropriate storage conditions; which it is typically stored at 4-6ºC in retailers and/or frozen in supermarkets [6]. However, the effect of different storage conditions on the textural properties, one of the most important quality parameters of phyllo, is still not well known. In this study, the effects of different storage temperatures and periods on the textural properties of phyllo were investigated. Phyllo samples were also examined in terms of color and microbiological properties during the storage period.

2. Material and Methods

2.1. Material

Flour (Çevikler Altınyazma) and salt used in phyllo production were obtained from Erzincan local market. Drinking tap water was used in preparing the phyllo. The properties of the flour were as follows; moisture content: 13.0%, wet gluten: 29.70%, dry gluten: 10.41%, gluten index: 92.75%, falling number: 1.035 s, Zeleny sedimentation: 35 mL. The L*, a* and b* color values of the flour were 85.50, -0.57 and 9.13, respectively. The farinograph parameters were as follows; water absorption: 69.1%, stability: 17.2 min., development time: 15.8 min., degree of softening: 35 FU. The extensograph parameters were as follows; extensibility: 114 mm, maximum resistance: 682 BU, energy: 91 cm².

2.2. Methods

2.2.1. Phyllo Production

Phyllo production was carried out by İrem Yufka company operating in Erzincan province (Figure 1). Firstly, 50 kg of flour was sifted, and 30 liters of water and 2 kg of salt were added. The mixture was kneaded for 30 min., the obtained dough was rested for 30 min., and then divided into balls of 130 g in size. After they were rolled out to a diameter of 60-70 cm, they were placed on a hot sheet and baked for 15 seconds on both sides. Then, they were soaked and rested for 2 hours. The rested phyllo samples were packaged in polyethylene bags with low moisture permeability and stored for short, medium and long periods in different storage conditions (3, 5 and 7 days at room temperature (20°C); 7, 14 and 21 days in refrigerator (4°C); 20, 40 and 60 days in deep freezer (-18°C)). The non-stored sample, i.e. the first day of production, was considered as the control sample.

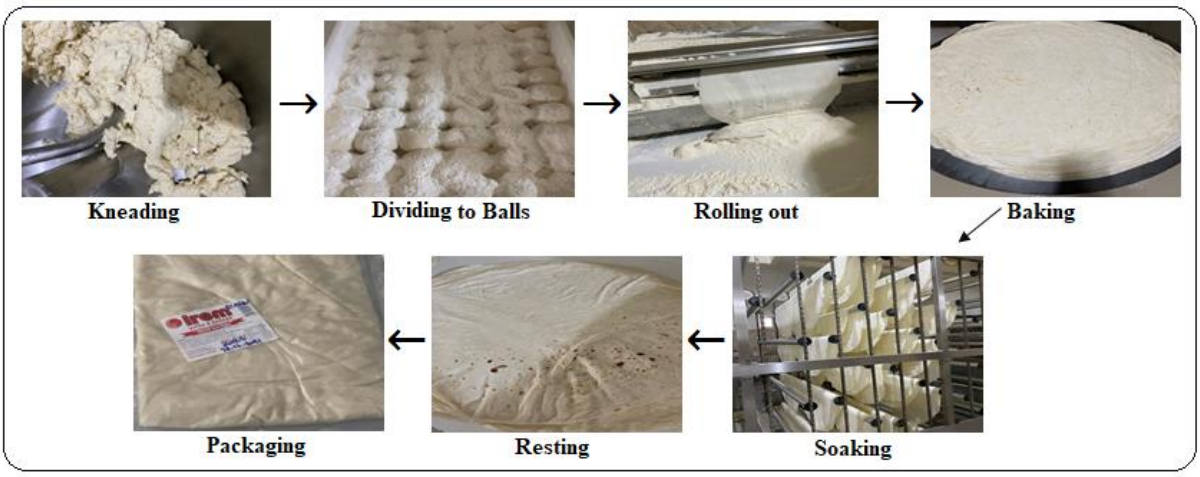

Figure 1. Phyllo production flow chart

2.2.2. Analysis of Phyllo Samples

2.2.2.1. Color Measurement

The color measurement of phyllo samples was carried out using the color analyzer (Minolta, CR-200, Japan) and measuring L*, a* and b* color values. Total color change (ΔE) was calculated as follows:

$$\Delta E = [(L_0-L_1)^2 + (a_0-a_1)^2 + (b_0-b_1)^2]^{1/2}$$

where, L_0, a_0, b_0 are L*, a* and b* color values of control phyllo sample (non-stored sample; first day of production), and L_1, a_1, b_1 are L*, a* and b* color values of stored phyllo samples, respectively.

2.2.2.2. Texture Profile Analysis (TPA)

Texture profile analysis in the phyllo samples (25 mm diameter, 4 layers (4x1 mm thickness)) was performed using a texture analyzer (TA.XTplus, Stable Micro Systems Ltd, Godalming, Surrey, U.K.) equipped with a 5 kg load cell and cylindrical metal probe (36 mm) (P/36). TPA test was carried out under conditions: 0.5 mm/s pre-test speed, 0.2 mm/s test and post-test speed, 30% compression rate, 20 g trigger force. The obtained results were expressed as hardness (N), cohesiveness, springiness, chewiness (N) and gumminess (N).

2.2.2.3. Stress Relaxation Test

The stress relaxation test was performed on phyllo samples with a texture analyzer (TA.XTplus, Stable Micro Systems Ltd, Godalming, Surrey, U.K.) equipped with Tortilla/Pastry Burst Rig under the following conditions: pre-test and test speed: 1 mm/s; post-test speed: 5 mm/s; holding time: 120 s; distance: 5 mm; trigger force: 5 g. Maximum force (g), minimum force (g) and relaxation time (s) values (time required for the maximum force to decrease to its 80%) were calculated from the obtained curve.

2.2.2.4. Stretching Test

The stretching test was performed on phyllo samples with a texture analyzer (TA.XTplus, Stable Micro Systems Ltd, Godalming, Surrey, U.K.) equipped with Tortilla/Pastry Burst Rig under the following conditions: pre-test and test speed: 1 mm/s; post-test speed: 10 mm/s; distance: 30 mm; trigger force: 5 g. Breaking force (g), breaking length (mm) and deformation modulus (g/mm) (breaking force/breaking length) were calculated from the obtained curve.

2.2.2.5. Microbiological Analysis

For the microbiological analysis, 90 ml of sterile physiological saline solution (0.85% NaCl) was added to 10 g of the sample and homogenized for 50 seconds. Plate Count Agar (PCA) and spreading method were used to determine total mesophilic aerobic bacteria count (TMAB). Petri dishes were incubated at 30°C for 48 hours. To determine total number of yeast and mold, Potato Dextrose Agar (PDA) and spreading method were used. Incubation was carried out at

28-30°C for 120 hours. The results were expressed as log colony forming units/g (log cfu/g) [11].

2.2.3. Statistical Analysis

All analyzes were carried out in triplicate. The data obtained were subjected to variance analysis using IBM® SPSS Statistics software version 22.0.0.0. The averages of main variation sources found to be important ($p<0.05$) were compared with the Duncan Multiple Comparison Test. The results were expressed as mean ± standard error. Pearson's Correlation Test was applied to determine the correlation among textural characteristics of phyllo samples.

3. Results and Discussion

3.1. Results of Color Measurement

Color is one of the most important criteria for food products [12]. Color, a physical characteristic that customers use to assess product quality [13], indicates the composition and freshness of the food products and provides information about whether there is falsification or not [12]. L*, a* and b* color values and total color change (ΔE) of phyllo samples are given in Table 1. L* value indicates the dark-light density of a product, and the closer to 100, the lighter the color [14]. Storage temperature significantly affected the L* color values of the samples ($p<0.01$). In general, the phyllo samples stored in the deep freezer exhibited higher L* color values, followed by the samples stored in the refrigerator and samples stored at room temperature, respectively. It is expressed that color change in the baked product originates from Maillard reactions which occur between proteins and reducing sugar and the increase in melanoidin concentration resulting from these reactions gives brownness to the product [15]. As known, the increasing the temperature causes the rate of the Maillard reaction to increase [16]. This situation explains why the highest level of L*color value was measured in the deep freezer temperature and the lowest level at room temperature. Although there was no statistically significant difference between the samples belonging to the short and medium period storage process in terms of the L* color value, an increase in storage time led to an increase in the L* color values of the samples stored at room temperature. Similarly, an increase was observed in the L* color values of the samples stored in the refrigerator due to the increase in the storage time. In the samples stored in the deep freezer, the increase in the storage time firstly increased the L* color value of the phyllo samples and then decreased this value. Nevertheless, the L* color values of the samples stored under deep freezer conditions were statistically close to each other and higher than the control sample in all three storage periods. It is thought that the degradation of color pigments over time may be effective in the increase in L* color values depending on storage time. On the other hand, it is known that color has a crucial importance on the acceptability of bakery products involving dough-based preparations, and a bright and clear appearance is generally desired [9]. Therefore, it can be said that the storage process and increasing the storage time in the refrigerator and deep freezer conditions provided positive changes in the phyllo samples in terms of L* color value, since its increase indicates increased brightness and clarity as desired.

Table 1. L*, a* and b* color values and total color change (ΔE) of phyllo samples[a]

Storage Temperature	Storage Time (Days)	L*	a*	b*	ΔE
	0 (Control)	77.61±0.33d	-0.53±0.03b	16.86±0.07a	-
	3	75.74±0.19e	-1.05±0.00f	13.42±0.12e	3.96±0.01bc
20ºC	5	76.60±0.39e	-0.94±0.01d	13.73±0.46e	3.33±0.55cd
	7	77.75±0.75d	-1.23±0.01g	14.12±0.02de	2.93±0.06cd
	GA	76.93±0.35C**	-0.94±0.10B**	14.53±0.52C**	3.40± 0.24B**
	7	78.53±0.14d	-1.34±0.01h	13.96±0.91e	3.16±0.88cd
4ºC	14	79.70±0.19c	-0.86±0.03c	16.41±0.07ab	2.17±0.21d
	21	81.75±0.18b	-0.43±0.00a	14.47±0.03cde	4.79±0.14ab
	GA	79.40±0.59B**	-0.79±0.13A**	15.43±0.50B**	3.37±0.54B**
	20	82.39±0.07ab	-1.34±0.00h	15.07±0.05cd	5.17±0.09ab
-18ºC	40	82.97±0.02a	-1.00±0.00e	16.58±0.02ab	5.39±0.02a
	60	82.33±0.01ab	-1.06±0.03f	15.54±0.03bc	4.94±0.01ab
	GA	81.33±0.82A**	-0.98±0.11C**	16.01±0.28A**	5.17±0.09A**
	P	**	**	**	**

[a] Means with different lowercase and uppercase letters in the same column are statistically different ($p < 0.05$).
**$P < 0.01$, GA: General Average.

a* value indicates the red/green color density of a product (+a: red, -a: green) [14]. The a* color values generally decreased in the stored samples compared to the control sample. Storage temperature affected the a* color values of the samples at a statistically significant level ($p<0.01$). In general, the phyllo samples stored in the refrigerator had higher a* color values, while the samples stored in the deep freezer had lower a* color values. When the storage period reached the middle period, a* color values of the phyllo samples increased in all storage temperatures. On the contrary, further increase in the storage time, which means that the storage time reached the long period, caused these values to decrease again in the samples stored at room temperature and in the deep freezer. However, in the samples stored in the refrigerator, a* color value increased when the long period was reached. b* value indicates the yellow/blue color density (+b: yellow, -b: blue) [14]. The b* color values generally decreased in the stored samples compared to the control sample. It was determined that the storage temperature affected the b* color values of the samples at a statistically significant level ($p<0.01$). In general, the phyllo samples stored in the deep freezer had higher b* color values, followed by the samples stored in the refrigerator and samples stored at room temperature, respectively. Although it was not statistically significant, the increasing the storage time caused an increase in b* color values of the samples stored at room temperature. In the samples stored in the refrigerator and deep freezer, medium period storage process caused an increase in b* color values, while the long period storage process caused a decrease.

It was determined that a statistically similar and lower total color change (ΔE) occurred in the phyllo samples stored at room temperature and in the refrigerator compared to the samples stored in the deep freezer. It is known that pigments can ordinarily degrade easily due to the many factors such as temperature, pH, light, etc. [17], however, they can be preserved at low temperatures away from these factors [18]. On the other hand, some studies have reported that exposure to low temperatures in different food samples can result in pigment damages such as

carotenoid degradation [19], decrease in chlorophyll content [20], etc. Therefore, it is thought that in this study, storage process in the deep freezer may have caused relatively more damage to the color pigments of the samples due to the low temperature effect, and therefore caused more total color change and variations in a* and b* values. In the samples stored at room temperature, the increasing the storage time decreased the total color change. In the samples stored in the refrigerator, medium period storage process caused a decrease in the total color change, while the long period storage process caused an increase. The opposite effect was observed in the samples stored in the deep freezer. That is, medium period storage process caused an increase in the total color change of the samples, while the long period storage process caused a decrease, however, all three values were statistically close to each other, especially the samples stored for short and long periods. When the color measurement parameters were examined in general and natural color of the phyllo was taken into consideration, it was concluded that although the total color change reached its maximum level, the deep freezer conditions, in which the highest L* color values were obtained, were better in terms of color properties, since a bright and clear appearance is desired [9].

3.2. Results of Texture Profile Analysis

The textural properties are very important for phyllo, especially in terms of quality of final product to be prepared using phyllo. The phyllo must have good flexibility and springiness properties. In addition, it must have good extensibility and be neither sticky nor weak enough to tear. All these features are desirable to supply it the ability to fold and roll during processing [2, 4, 10]. Hardness, cohesiveness and springiness values of phyllo samples are given in Figure 2. Hardness is the force required to deform a sample to a certain level [21]. Both very hard and very soft structures are undesirable in the phyllo as it affects the quality of the final product to be prepared using phyllo. The hardness values generally decreased in the stored samples compared to the control sample. The decrease in the hardness values of the samples due to the storage process was probably related to amylolytic and proteolytic enzymes present in the flour, which were activated by water during dough preparation and originated from microbial activity (Table 5), and phase transformations of water. In general, the phyllo samples stored in the refrigerator exhibited higher hardness values, followed by the samples stored at room temperature and samples stored in the deep freezer, respectively. The freezing process is one of operation that significantly affects dough quality [22]. During the freezing process, water molecules bond with hydrogen bonds to form a hexagonal crystal structure, leading the volume increases by approximately 9% [23]. The formed ice crystals negatively affect especially proteins in the dough system, as a result, three-dimensional gluten network is damaged by ice crystals [22]. Moreover, when thawing, which is necessary for rehydration of the gluten matrix in the frozen dough, is carried out at a certain temperature, condensation phenomenon takes place on the dough surface because the dough has a lower temperature than surrounding air [22], that is, the dough cannot regain all the water it lost. All these cases lead to significant decreases in the strength of dough [22] and, therefore the hardness of samples stored in the deep freezer decreases further. In addition, it is thought that the reason why the samples stored at room temperature had lower hardness values than the samples stored in the refrigerator may primarily due to higher enzyme activity. As it is known that enzymes have optimal temperature

requirements for maximum activity. Most of the enzymes work optimally between 30-70°C [24]. The low temperature ranges do not generally meet the needed activation energy, leading to the decreases in enzyme activity [25]. The increasing the storage time caused a significant decrease in the hardness values of the samples stored at room temperature and in the deep freezer. This decrease appears to be due to a general increase in the microbial activity linked to enzyme activity (Table 5). Although there was no statistically significant difference between medium and long period storage processes, the increase in the storage time had an increasing effect on the hardness values of the samples stored in the refrigerator. This may be due to the environment being relatively unfavorable in terms of temperature for enzymes activity. In addition, it is thought that the increase in the hardness values of the samples stored in the refrigerator depending on the storage time may have caused by partial retrogradation. It is mainly responsible for the hardness that occurs over time in foods containing starch [26] and occurs either too slow or to dense at extreme temperatures due to Brownian motion mechanism of macromolecules [27], which this range, where the highest hardness was measured, was a relatively medium temperature degree in this study.

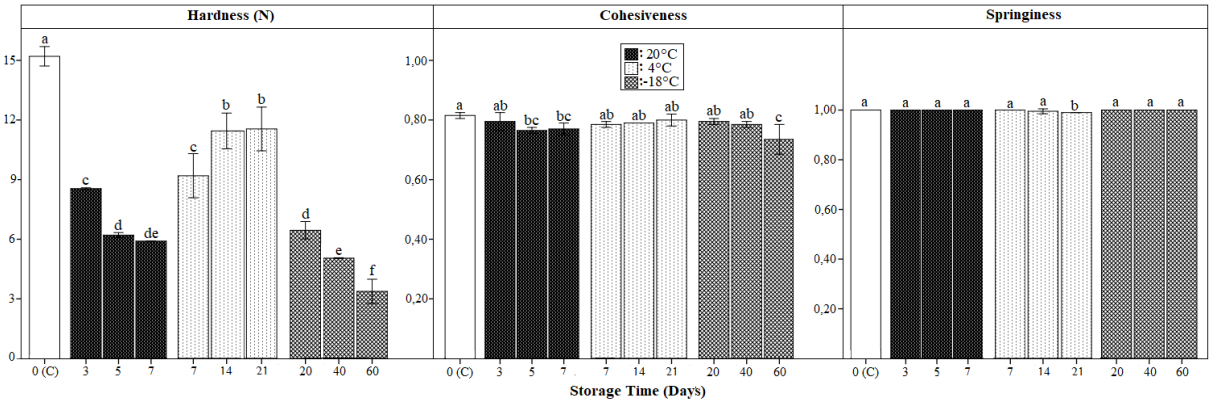

Figure 2. The effect of different storage temperatures and periods on the hardness, cohesiveness and springiness values of phyllo samples, respectively (C: Control)

Cohesiveness is described as the density of internal bonds in the structure of food [28]. It was determined that the storage process caused the phyllo samples to tend to exhibit lower cohesiveness values in general. The samples stored in the refrigerator had higher cohesiveness values, followed by the samples stored at room temperature and samples stored in the deep freezer in general, however, it was not statistically significant. The increasing the storage time had a reducing effect on the cohesiveness values of the samples stored at room temperature and in the deep freezer in general. The opposite effect was observed in the samples stored in the refrigerator, but it was not statistically significant. It was determined that the changes in the cohesiveness values of the samples depending on the storage time and temperature were similar to the changes in the hardness values in general. Therefore, it is thought that the occurrences that damage the hardness properties of samples such as breakdown of proteins, which gluten is mainly responsible for cohesiveness of bakery products [29], and carbohydrates due to microbial and enzyme activity, ice crystal formation [22] and failure to completely rehydration during thawing caused similar injuries, weakening the internal structure of the samples and leading to loss of dough strength. It was already determined that there was a significant correlation between the hardness and cohesiveness values of the phyllo samples ($r = 0.802^{**}$)

(Table 4). In addition, it is expressed that the decrease in the cohesiveness due to the increase in the storage time is caused by instability of protein macromolecular colloid originated from the freezing concentration effect such as changes in solubility and denaturation of protein and cohesion force among the protein molecules, and this case tends to the increase with the increase in the storage time, as seen in deep freezer storage process in this study. Supportively our findings, it is stated that longer frozen storage periods have also an unfavorable effect on interactions among starch and gluten network [30].

Springiness is an expression of how well a sample springs after initial deformation [31]. As mentioned earlier, phyllo must have a good springiness to obtain optimum processing properties. It was determined that the samples stored at room temperature and in the deep freezer exhibited statistically similar and higher values than the samples stored in the refrigerator. Only the sample stored in the refrigerator for 21 days exhibited a statistically lower springiness value than the other samples. Nevertheless, there was no numerically sharp decrease in the springiness value of this sample, meaning that all phyllo samples had similar springiness properties to the control group in general. It can be said that the fact that different storage conditions, including long periods and wide temperature ranges, do not significantly affect the springiness property, which is essential for phyllo, is a quite positive and desirable result for this study.

Table 2. Chewiness and gumminess values of phyllo samples[a]

Storage Temperature	Storage Time (Days)	Chewiness (N)	Gumminess (N)
	0 (Control)	12.37±0.22a	12.39±0.22a
	3	6.77±0.13c	6.80±0.14c
20ºC	5	4.71±0.01de	4.72±0.00de
	7	4.45±0.00ef	4.43±0.03ef
	GA	7.08± 1.21B**	7.09± 1.21B**
	7	7.19±0.39c	7.22±0.41c
4ºC	14	9.75±0.25b	9.75±0.25b
	21	9.90±0.30b	9.88±0.31b
	GA	9.80±0.70A**	9.81±0.70A**
	20	5.13±0.14d	5.14±0.14d
-18ºC	40	3.94±0.02f	3.95±0.02f
	60	2.47±0.14g	2.48±0.14g
	GA	5.98±1.44C**	5.99±1.44C**
	P	**	**

[a] Means with different lowercase and uppercase letters in the same column are statistically different ($p < 0.05$).
**$P < 0.01$, GA: General Average.

Chewiness and gumminess values of phyllo samples are given in Table 2. Generally, both chewiness and gumminess decreased in the stored samples compared to the control sample. It was determined that the storage temperature affected the chewiness and gumminess values of the samples at a statistically significant level ($p<0.01$). In general, the phyllo samples stored in the refrigerator had higher chewiness and gumminess values, while the samples stored in the deep freezer had lower values. In the samples stored at room temperature and in the deep freezer, the increase in the storage time decreased the chewiness and gumminess values. In the

samples stored in the refrigerator, chewiness and gumminess values increased with increasing the storage time, however, there was no statistically significant difference between medium and long storage periods. Chewiness is defined as the measure of energy required to make solid foods ready to be swallowed while being chewed in the mouth [32]. It is calculated by multiplying the hardness, cohesiveness and springiness parameters. Gumminess is the measure of the energy required to make semi-solid foods ready to swallow. It is calculated by multiplying the hardness and cohesiveness parameters [21]. Chewiness and gumminess are secondary textural parameters derived from primary textural attributes. Consequently, these parameters are influenced by fundamental factors, particularly hardness, as the hardening of food texture typically indicates an increase in the energy required to make the food ready for swallowing. Therefore, the changes in the hardness values of the phyllo samples, depending on storage temperature and duration, were directly reflected in the chewiness and gumminess values. Consistently, positive and significant correlations were observed between hardness and chewiness ($r = 0.997^{**}$), hardness and gumminess ($r = 0.997^{**}$), and chewiness and gumminess ($r = 1.000^{**}$) (Table 4).

3.3. Results of Stress Relaxation Test

The stress relaxation test, used to investigate the viscoelastic properties of foodstuffs, is based on measuring the force required to maintain deformation under a certain stress value over time [33]. Relaxation time, maximum force and minimum force values of phyllo samples, that are the basic textural parameters obtained from the stress relaxation test, are given in Table 3. Relaxation time value expresses to the time required for the maximum force to decrease to its 80% for this study. It was determined that the storage temperature affected the relaxation time values of the phyllo samples at a statistically significant level ($p<0.01$). The samples stored at room temperature and in the deep freezer exhibited statistically similar and lower relaxation time values than the samples stored in the refrigerator. As the storage time increased, the relaxation time value firstly decreased and then increased in the samples stored at room temperature and in the deep freezer. However, they could not reach their levels in the short period storage time, in which case it can be said that the increase in the storage time had a generally decreasing effect on the relaxation time values of these samples. In the samples stored in the refrigerator, the increasing the storage time caused relaxation time values to increase. It is thought that the changes in the relaxation time values of the stored samples due to temperature and period variation sources may be caused by changes in the hardness properties of the samples. The lower relaxation time is associated with the material that shows liquid-like property, on the contrary, the higher relaxation time is associated with a more solid property [34]. Based on this, it is thought that the samples stored in the refrigerator, which had higher hardness values (Figure 2), exhibited higher relaxation time values. In addition, the samples stored at room temperature and in the deep freezer may have exhibited close and lower relaxation time values, since they had close and lower hardness values than the samples stored in the refrigerator even if statistically different. Similarly, as storage time increased, the relaxation time values of the samples stored at room temperature and in the deep freezer generally decreased, while the relaxation time values of the samples stored in the refrigerator increased, which a similar change trend was also observed in the hardness values of the samples.

Table 3. Relaxation time, maximum force and minimum force values of phyllo samples[a]

Storage Temperature	Storage Time (Days)	Relaxation Time (s)	Maximum Force (g)	Minimum Force (g)
	0 (Control)	8.63±0.22d	33.83±0.05f	24.74±0.12f
	3	11.06±0.00b	41.02±0.50c	33.52±0.49c
20ºC	5	8.68±0.02cd	37.71±0.37e	31.92±0.33d
	7	9.53±0.13bcd	39.38±0.28d	30.66±0.66d
	GA	9.48±0.37B**	37.99± 1.02B**	30.21± 1.26B**
	7	8.48±0.06d	42.30±0.20b	36.55±0.55b
4ºC	14	9.29±0.52cd	43.17±0.18b	37.35±0.55b
	21	17.61±1.28a	50.98±0.79a	41.46±0.50a
	GA	11.00±1.47A**	42.57±2.30A**	35.03±2.36A**
	20	11.01±0.32b	32.10±0.10g	25.35±0.25ef
-18ºC	40	9.71±0.00bcd	31.60±0.40g	26.37±0.14e
	60	10.38±0.61bc	31.45±0.45g	25.80±0.56ef
	GA	9.93±0.36B**	32.24±0.38C**	25.57±0.26C**
	P	**	**	**

[a] Means with different lowercase and uppercase letters in the same column are statistically different ($p < 0.05$).
**$P < 0.01$, GA: General Average.

It was determined that the storage temperature significantly affected the maximum force values of the phyllo samples ($p<0.01$). While the samples stored at room temperature and in the refrigerator showed higher maximum force values than the control sample, the samples stored in the deep freezer showed lower values. Among the samples subjected to storage process, the samples stored in the refrigerator exhibited higher maximum force values, followed by the samples stored at room temperature and samples stored in the deep freezer, respectively. For the samples stored at room temperature, the increasing the storage time initially decreased and then increased the maximum force values, though there was a general trend of decrease. Although there is no statistically significant difference between the short and medium period storage process, the increase in the storage time had an increasing effect on the maximum force values of samples stored in the refrigerator. It had also a decreasing effect on the maximum force values of samples stored in the deep freezer, however, it was not statistically significant. The storage process was generally effective in increasing the minimum force values of the phyllo samples compared to the control sample. The storage temperature affected the minimum force values of samples at a statistically significant level ($p<0.01$). In general, the samples stored in the refrigerator had higher minimum force values, while the samples stored in the deep freezer had lower values. Although there was no statistically significant difference between the samples stored in the medium and long period, the increasing the storage time caused a decrease in the minimum force values of samples stored at room temperature. In the samples stored in the refrigerator, the increasing the storage time caused an increase in the minimum force values of samples, however, there was no statistically significant difference between the samples stored in short and medium period. In the samples stored in the deep freezer, the minimum force value firstly increased and then decreased with the increase in the storage time, however, the minimum force values in all three storage periods were statistically similar. Similar to relaxation time, the trends in maximum and minimum force values can also be attributed to changes in the hardness of the samples. The maximum force refers to the measure of the initial resistance of

the sample to deformation in the course of stress relaxation analysis, while the minimum force refers to the measure of the ultimate resistance measured at the end of the analysis. The increase in mentioned values, which are closely connected with the hardness of food structure, implies that the material shows a more solid-like property and requires higher deformation amount [35]. For this reason, the changes in hardness values of the samples depending on storage temperature and period were generally reflected in maximum and minimum force values (Figure 2).

3.4. Results of Stretching Test

The effects of different storage temperatures and periods on the breaking force, breaking length and deformation modulus values of phyllo samples are given in Figure 3, respectively. It was determined that the samples stored in the deep freezer exhibited higher breaking force values, while those stored in the refrigerator generally showed lower values. As the storage time increased, the breaking force values of the samples stored at room temperature decreased significantly. A general decrease was also observed in the samples stored in the deep freezer, however, the breaking force values of the samples stored medium and long period were statistically insignificant. In the samples stored in the refrigerator, when the storage period arrived to the middle period, the breaking force value increased, but further increase decreased this value again. It was determined that the breaking force values of the phyllo samples had a negatively correlation with the maximum force ($r = -0.645^{*}$) and minimum force ($r = -0.657^{*}$) values (Table 4), suggesting that changes in breaking force values of the samples may depend on this relationship. That is, the samples with higher maximum and minimum force values, which was previously stated to indicate a harder sample structure, exhibited easier breakable property due to their harder structure, and this situation caused them to exhibit lower breaking force values. As another pointer, it is clearly seen in Figure 2 that the samples with higher hardness values had lower breaking force values, while the samples with lower hardness values had higher breaking force values when comparing on the group basis.

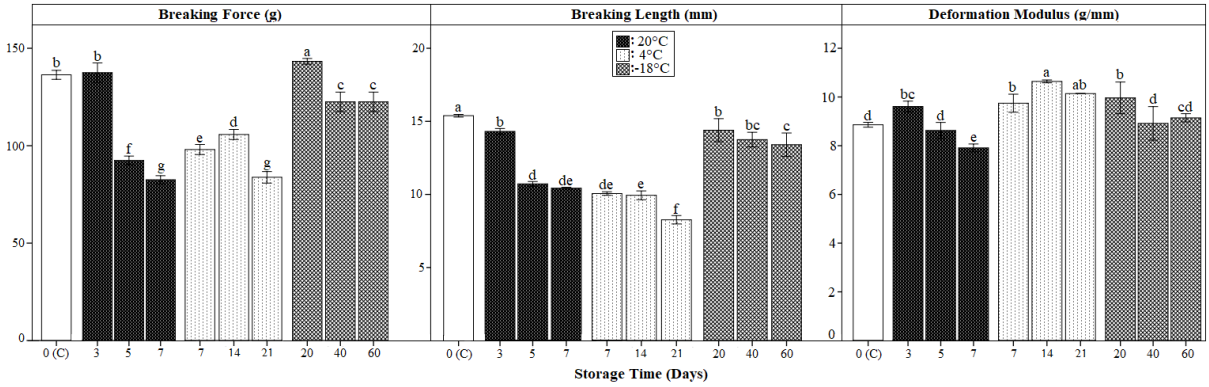

Figure 3. The effect of different storage temperatures and periods on the breaking force, breaking length and deformation modulus values of phyllo samples, respectively (C: Control)

Breaking length refers to the average distance required for the breaking process, and the increasing this distance means that the dough extensibility increases [36]. It was determined that the storage process caused the breaking length values of all samples to decrease, which expresses that the control sample had the highest breaking length value. In general, the samples stored in the deep freezer exhibited higher breaking length values, followed by the samples

stored at room temperature and samples stored in the refrigerator, respectively. At all three storage temperatures, there was a general decrease in the breaking length values of the samples in parallel with the increase in the storage time. The changes in the breaking length values of the samples can be generally attributed to the close relationship between breaking length and breaking force values ($r = 0.926^{**}$) (Table 4). As stated in the previous section, the decrease in the breaking force values of the samples is due to the increase in the breaking ability resulting from the increase in the hardness of the sample structure, facilitating the breaking action. That is, the decrease in the breaking force means that the breaking action ends faster, leading to a decrease in the breaking length value expressing the distance required for breaking. Therefore, the samples with lower breaking force values exhibited lower breaking length values, while the samples with higher breaking force values exhibited higher breaking length values. For these reasons, phyllo samples with higher springiness, which implies that they can tolerate the applied deformation better and do not immediately deform/rupture/break, exhibited higher breaking length values ($r = 0.656^{*}$) (Table 4).

It was determined that the samples stored in the refrigerator had higher deformation modulus values, followed by the samples stored in the deep freezer and samples stored at room temperature, respectively. As the storage time increased, the deformation modulus values generally decreased in the samples stored at room temperature and in the deep freezer. In the samples stored in the refrigerator, the deformation modulus values firstly increased and then decreased to a similar statistical level depending on the storage time. Deformation modulus is a parameter obtained by using the breaking force and breaking length values, stating that it is characterized by these values, and gives an idea about the processability properties of the phyllo. When the stretching/tearing properties of phyllo are considered, the deformation modulus is desired to be low, while the breaking force and breaking length values are desired to be high, which enables that the phyllo is not too weak to tear in order to obtain optimum processing performance. In this context, it can be said that storage process in the deep freezer conditions was generally more suitable, as the samples stored in that condition had relatively higher breaking force and breaking length values with a moderate deformation modulus.

Table 4. Pearson's correlation coefficient of textural characteristics of phyllo samples

	Hardness	Cohesiveness	Springiness	Chewiness	Gumminess	Max. Force	Min. Force	Relaxation Time	Breaking Force	Breaking Length
Cohesiveness	0.802**	-	-	-	-	-	-	-	-	-
Springiness	-0.441	-0.292	-	-	-	-	-	-	-	-
Chewiness	0.997**	0.799**	-0.500	-	-	-	-	-	-	-
Gumminess	0.997**	0.800**	-0.497	1.000**	-	-	-	-	-	-
Maximum Force	0.465	0.304	-0.783**	0.493	0.491	-	-	-	-	-
Minimum Force	0.350	0.190	-0.738*	0.381	0.380	0.968**	-	-	-	-
Relaxation Time	0.150	0.207	-0.809**	0.199	0.197	0.583	0.489	-	-	-
Breaking Force	0.025	0.253	0.460	0.010	0.013	-0.645*	-0.657*	-0.226	-	-
Breaking Length	-0.088	0.128	0.656*	-0.119	-0.116	-0.801**	-0.844**	-0.367	0.926**	-
Deformation Modulus	0.376	0.362	-0.577	0.421	0.422	0.437	0.512	0.391	0.165	-0.216

*p<0.05 ** p<0.01

3.5. Results of Microbiological Analysis

Total mesophilic aerobic bacteria (TMAB), and yeast and mold count of phyllo samples are given in Table 5. Compared to the control sample, storage process at room temperature and in

the refrigerator caused an increase in the total mesophilic aerobic bacteria count of the samples, while the deep freezer conditions caused a decrease. In general, the samples stored at room temperature had higher total mesophilic aerobic bacteria count, followed by the samples stored in the refrigerator and deep freezer, respectively. As it is known, microorganisms have certain temperature requirements for growth and reproduction, and each microorganism has optimum temperature ranges in which it reproduces at the maximum level [37]. In general, mesophilic bacteria reproduce at 37ºC at an optimum level [38], while molds and yeasts reproduce at 25-30ºC [37]. Reproduction of microorganisms decreases as environment temperature moves away from these limits [39] as in this study. As expected, the increasing the storage time at all three storage temperatures generally led to an increase in the total mesophilic aerobic bacteria numbers of the samples and it was especially higher at room temperature. The change trend in the yeast and mold count was partially similar for similar reasons. Storage process at room temperature and in the refrigerator caused an increase in the total yeast and mold count of the phyllo samples compared to the control sample. An increase was also observed in the samples stored in the deep freezer conditions, but, it was relatively lower. The increasing the storage time resulted in an increase in the total number of yeast and mold in the samples stored at room temperature and in the refrigerator. In the samples stored in the deep freezer, the increase in the storage time firstly increased and then decreased the total number of yeast and mold. The increase in microbial load due to prolonged storage time, which is one of the stages that should be given importance in terms of microbiological stability [40], has also been observed in different food samples [41]. Considering the effect of the storage process on the microbial load of the phyllo samples, it was concluded that the storage process in the deep freezer conditions was more promising due to a relatively lower total mesophilic aerobic bacteria and mold and yeast count with a relatively lower overall rate of increase.

Table 5. Total mesophilic aerobic bacteria (TMAB) and yeast and mold count of phyllo samples

Storage Temperature	Storage Time (Days)	TMAB (log cfu/g)	Total Number of Yeast and Mold (log cfu/g)
	0 (Control)	6.61	<2.00
	3	7.94	4.72
20ºC	5	9.81	6.41
	7	12.30	6.65
	7	8.69	3.79
4ºC	14	8.38	3.98
	21	8.98	6.87
	20	3.38	<2.00
-18ºC	40	4.27	3.04
	60	4.43	2.69

4. Conclusion

Phyllo is an important cereal-based semi-product. Phyllo, which has an important role in human nutrition, is generally stored in cold storage conditions due to its short shelf life. In this study, the effect of different storage conditions on the textural, microbiological and color properties of phyllo was investigated. In general, the phyllo samples stored in the deep freezer exhibited higher L* color values, followed by the samples stored in the refrigerator and samples stored at

room temperature, respectively. A statistically similar and lower total color change (ΔE) occurred in the phyllo samples stored at room temperature and in the refrigerator than the samples stored in the deep freezer. The samples stored in the refrigerator exhibited higher hardness values, followed by the samples stored at room temperature and samples stored in the deep freezer, respectively. The storage temperature did not affect statistically significantly the cohesiveness values of the samples. Most of the phyllo samples were similar to the control sample in terms of springiness property. The samples stored in the refrigerator had higher chewiness and gumminess values, while the samples stored in the deep freezer had lower values. In terms of relaxation time value, the samples stored at room temperature and in the deep freezer exhibited statistically similar and lower values than the samples stored in the refrigerator. In addition, the samples stored in the deep freezer generally had higher values in terms of the breaking force and breaking length. For microbiological properties, the storage process in the deep freezer conditions was more appropriate by reason of a relatively lower total mesophilic aerobic bacteria and yeast and mold count with a relatively lower overall rate of increase.

As a result, it was concluded that the deep freezer conditions were more suitable for preserving the color and microbial properties of phyllo. Considering textural properties of phyllo, it was determined that the refrigerator conditions were more suitable in terms of hardness, chewiness, gumminess, relaxation time, maximum force and minimum force values, while the deep freezer conditions were more suitable in terms of springiness, breaking force and breaking length. Overall, considering the textural quality of phyllo, it would be more appropriate to recommend deep freezer conditions, as the springiness and stretching properties of phyllo are relatively more important in terms of processability.

Ethics in Publishing

There are no ethical issues regarding the publication of this study.

Author Contributions

This article is a part of Seda COŞKUN's Master's Thesis titled "Effects of different storage conditions on textural and microbiological properties of yufka" under the supervisor of Prof. Dr. M. Murat KARAOĞLU. Aslıhan Hanoğlu and Yeşim Bedir contributed to the laboratory studies of this study and the writing of this article.

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