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The Effects of Two-Step Tempering Treatment on the Physical, Chemical and Technological Properties of Flour in Bread Wheats (*Triticum aestivum* L.)

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ABSTRACT

In this study, the effects of two-step tempering treatment on the physical, chemical and physicochemical characteristics of bread wheat (*Triticum aestivum* L.) flour were investigated. Two wheat varieties (Adana-99 and Russian) with different variety properties were used in the research. Each wheat variety was subjected to four different tempering treatments. These treatments were; a. no tempering (control), b. single-step tempering for 24 h, c. single-step tempering for 48 h and d. two-step tempering for 48 h. Following the treatment procedures, flour samples obtained from wheat samples fed to the mill were subjected to chemical and technological analyses two days after the flour samples were obtained. Results indicated that the tempering treatment had a limited improvement in the physical (flour yield and color) and chemical properties of the flour. It was found that, the tempering treatment reduced the ash and protein contents of the flour samples, however it resulted in an increase in the gluten quality. Tempering treatment for 48 h among single-step tempering treatments resulted in more improved flour quality. The findings conclude that, when water added to the wheat in the tempering process in two steps results in an improved flour quality. However, further studies are needed to elucidate the mechanism.

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Ekmeklik Buğdaylara (*Triticum aestivum* L.) İki Aşamalı Uygulanan Tavlama İşleminin Unun Fiziksel, Kimyasal ve Teknolojik Özelliklerine Etkileri

ÖZET

Bu çalışmada; ekmeklik buğdaya (*Triticum aestivum* L.) iki aşamalı uygulanan tavlama işleminin buğday ununun fiziksel, kimyasal ve fizikokimyasal özellikleri üzerine etkileri araştırılmıştır. Araştırmada, çeşit özellikleri farklı iki buğday örneği (Adana-99 ve Rus) kullanılmıştır. Her bir buğday çeşidi tavlama ile ilgili dört ayrı muameleye tabi tutulmuştur. Bu uygulamalar; a. tavsız (kontrol), b. 24 saat süreyle tek aşamalı tavlama, c. 48 saat süreyle tek aşamalı tavlama ve d. 48 saat süreyle iki aşamalı tavlama. Söz konusu muameleler neticesinde değirmene beslenen buğday örneklerinden elde edilen un numuneleri iki gün sonra kimyasal ve teknolojik analizlere tabi tutulmuştur. Bulgular şu şekilde özetlenebilir: Tavlama muamelesi unun fiziksel (un randımanı ve renk) ve kimyasal özelliklerinde sınırlı iyileşme sağlamıştır. Yine, tavlama muamelesi un örneklerinin kül miktarında daha belirgin olmak üzere protein miktarında azalmaya, ancak gluten kalitesinde ise artışa yol açmıştır. Tek aşamalı tavlanmış örnekler arasında 48 saat süreyle tavlama un niteliklerini daha fazla geliştirmiştir. Bulgular, tavlama prosesinde buğdaya verilen su miktarının kademeli olarak iki defada verilmesinin un niteliklerini geliştirme noktasında daha iyi sonuç sağladığını ortaya koymuştur. Ancak, bu mekanizmayı aydınlatmak için daha fazla çalışmaya ihtiyaç vardır.

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INTRODUCTION

Cereals are the main food raw materials that people have consumed since ancient times. Wheat is a major raw material used in the production of many bakery products, especially bread, and has a unique place in cereals due to its unique characteristics, unlike other cereal flours (Dunn et al., 2015). In Turkey, as in many countries, wheat ranks first among cereals in terms of both cultivation area and production quantity (20.5 million tones according to 2020-year data; [Anonim, 2021]). it is an important culture plant due to its ease of cultivation, its adaptability of conversion to wide variety of foods and its role in nutrition.

Wheat grain is anatomically formed from outside to inside bran (13-17%), germ (2-3%) and endosperm layers (80-85%; [Delcour and Hoseney, 2010]). The aim of wheat milling is to separate the endosperm (flour or semolina) from the bran and germ layers. The processing of wheat in flour and semolina in milling can be grouped under three major groups: a) Preparation processes (buying and storing of wheat, cleaning, and separating it from foreign substances, blending, washing [if necessary], and tempering), b) Milling (grinding) operation (with the help of sieving tools and semolina-bran purification devices with crushing [breaking] and reducing rolls) and c) Flour storage and blending operations. All these process steps affect milling products' qualitative and quantitative properties (flour, semolina, starch, bran, germ, bon quality) to be obtained (Kent, 1984; Delcour and Hoseney, 2010). Additionally, the labor and the rigor shown in the preparation processes help obtain high-quality flour and semolina. Also, there is overall reduction in energy consumption of the milling factory and these results in roller-sieve systems to be used efficiently and an increased life span. For these reasons, preparation processes in milling are the processes that need to be considered. In these processes, tempering has a special and important place in cereal industry.

The optimum amount of water is supplied to the grain by means of the tempering process to make the physical characteristics of the wheat grain suitable for milling and to improve the milling quality. Than the wheat stack is rested in the silos for a certain time to equilibrate the moisture content and allow evenly distribution, which will result in optimum properties for milling (until the optimum moisture distribution and milling properties are reached for milling). The water supplied to the wheat by tempering, enters the grain through diffusion and spreads (Keskinoğlu et al., 2001). In this way, bran (shell) having a very rigid structure is broken more easily by achieving an elastic structure (the energy consumption of the milling factory is reduced) and the loosening of the bond between the bran and the endosperm layers of the grain, which are tightly bonded to each other, is easier to separate from the flour (Cornell and Hoveling, 1998; Yoo et al., 2009; MacRitchie, 2010). Tempering process; it should be such that it balances flour extraction and bran ratio (Hook et al., 1982; Kweon et al., 2009). Many factors such as grain hardness, initial moisture of wheat, time to be applied, lying time, amount and temperature of water, quality and quantity of intermediate product to be obtained are effective on tempering (Cornell and Hoveling, 1998; MacRitchie, 2010).

As is known, the quality of the bread made from the new milled flour is relatively low. These flours need to be rested for a while to improve their bread making performance. For the flour to mature under normal conditions, the flour should be rested for 3-4 weeks after the milling process. Flour maturing and bleaching chemical agents (such as chlorine gas, benzoyl peroxide, potassium bromate and azodicarbonamide) may be carried out within a few hours of desirable improvements. The flours treated in this way are less resistant to long-time storage than those not treated (Dizlek and Kurt, 2017). Although these chemicals provide serious advantages to the wheat milling factories according to the natural bleaching of the flour; flour storage area and saving time, they have the potential a risk / problem in terms of human health and thus limit the use of these substances today.

Tempering has a great effect, especially at the point of lowering the ash content of flour. Flour export in Turkey has an increasing momentum in recent years. At present, whiter flours with higher parity are preferred in flour exports to the Middle East from Turkey. Therefore, the importation of flour by wheat milling industry, which has started to make wheat importation and which provides value added by processing this wheat in particular flour and sometimes semolina, is the color of white matter which is emphasized by importers (Kurt and Dizlek, 2020). For this reason, in the study, an application was used (wheat has been tempered by two-steps) that can be important in terms of providing flour to support flour export. Thus - in the pilot scale – it is tried to produce high parity flour that has low color and ash values. Briefly, in the study it is aimed to bleach the flour with a natural method. The possible good results of this application may be an alternative to the use of chemicals which are frequently used in bleaching of flour color but which are risky. In addition, improper tempering; increases the volume of tempering silos, extends the tempering period, increases the number of mechanization units; increases investment, labor and maintenance costs and increases the energy and cost spent for grinding.

The rate of moisture movement into the kernels, tempering time, tempering water temperature and different tempering methods (warm, hot, steam, microwave, ultrasound etc.) have been emphasized in the previous studies (Butcher and Stenvert, 1973; Stenvert and Kingswood, 1976; Moss, 1977; Finney and Bolte, 1985; Ertugay et al., 1995a; 1995b; 1995c; Cornell and Hoveling, 1998; Ibanoglu, 2001; Keskinoğlu et al., 2002; Sünter, 2003; Kweon et al., 2009; Warechowska et al., 2016). The amount of water to be supplied to the wheat mass during tempering is usually given at single-step. However, in this study, two bread wheat (*Triticum aestivum* L.) varieties with different degrees of hardness were subjected to the two-step tempering process as well as classical (single-step) tempering process. In this way, the effect of the two-step tempering process applied to the bread wheat on the physical, chemical and technological properties of the flour was investigated.

MATERIAL and METHODS

Materials

Wheat: In the study, two different bread wheat (*Triticum aestivum* L.) varieties namely a low-protein, soft 'Russian' variety imported from Russia and a domestic, medium hard 'Adana-99' wheat variety were used. Both wheat samples are 2017 crop season; The Adana-99 variety was supplied from İslamoğlu Trade in Osmaniye (Turkey) and the Russian variety was supplied from Sunar Özlem Flour Factory (Osmaniye, Turkey). Approximately 100 kg of each wheat variety was supplied. The samples taken from the wheat masses in accordance with the sampling method were stored in 50 kg jute bags and stored in cold storage (+4 °C) until they were analyzed and ready for milling.

Flour: After applying different tempering processes to the two wheat varieties, the wheat varieties were milled according to method (Kurt and Dizlek, 2020) described in method section to obtain subsequent flours.

Water: Potable water supplied within the campus of University of Osmaniye Korkut Ata (Osmaniye, Turkey) was used in tempering process of the wheat samples.

Laboratory Type Mill: Wheat samples were milled into flour by using; the first three pieces of crushing (breaking), the last two reducing rolls, a total of five rolls of 'Chopin' brand 'CD1' model tempered wheat grinding mill (Chopin Technologies, Paris, France).

Formation of Experimental Wheat Groups

The target moisture content of the Adana-99 wheat used in the study was 17% and the Russian wheat was 16.5%. The amount of tempering water for given masses of wheat was calculated according to American Association of Cereal Chemists International (AACCI) Method 26-95.01 (AACCI, 2010). Wheat samples were tempered with cold tap water. Within the scope of this research, the experimental materials and the pattern were formed by applying the following different tempering treatments in each of the two bread wheat varieties whose qualities are different:

a) no tempering (= control)

b) single-step tempering for 24 h (The amount of water calculated according to the target moisture is given to the wheat at one time and tempered for 24 h)

c) single-step tempering for 48 h (The amount of water calculated according to the target moisture was given to the wheat in one time and tempered for 48 h)

d) two-step tempering for 48 h (Adana-99 and Russian wheat were initially treated with 15% and 15.5%, respectively, with the first tempering water and resting for 24 h. At the end of the 24 h period, second tempering water added to Adana-99 and Russian wheat to a target moisture of 17% and 16.5%, respectively (In summary, the wheat samples were tempered two times and the final moisture content of the wheat was adjusted to the percentage in b) and c)).

e) The flour obtained from wheat, (tempered according to item d)), was rested in cool and dry room conditions for 30 days (Wheat flour that has been tempered two-step for 48 h and kept in room conditions for 30 days).

Milling Process

Optimally tempered wheat samples were separately milled into flour using a laboratory mill. The milling consisted of two parts: crushing and reduction. Cleaned and tempered wheat was first broken by the crushing system, and then semolina, coarse bran and some crushed flour were separated from crushed wheat. The semolina obtained as the main product was reduced to the flour by milling in the reduction system and divided into refined bran by-products. The separated refined bran was passed through the reduction system for a second time to obtain whole flour (breaking flour + first reduction flour + second reduction flour). These flour samples were used for chemical and technological analyses. Flour samples obtained from crushing and reduction systems were weighed and the flour yields were calculated as % by proportioning to the amount of milled wheat that was to the initial cleaned and tempered wheat (Kurt and Dizlek, 2020).

Analysis of Wheat Samples

To determine the physical properties of wheat samples; the percentage of sunn pest damaged grain (Dizlek and Ozer, 2016a); hectoliter weight, thousand kernel weights, kernel vitreousness analyses were established according to procedure Uluöz (1965); in addition, flour yield was calculated. The chemical properties of wheat samples were also determined. For this purpose; moisture, crude protein (AACCI Method 39-25.01 [AACCI, 2010]) and starch contents of the wheat samples were determined by using NIR method (Near infrared reflection spectroscopy; NIR Analysis System, Perten Diode Array 7250, Stockholm, Swedish).

Analysis of Flour Samples

In this study, in order to determine some of the chemical properties of the flour samples obtained from five different tempering treatment methods; the amounts of ash (AACCI Method 08-01.01 [AACCI, 2010]), crude protein and moisture (AACCI Method 39-11.01 [AACCI, 2010]) and damaged starch (by the NIR method) were determined. Color values of flour samples were also determined. For determination of the physicochemical properties of flour samples; which are important for bread-making, wet and dry gluten amounts (with near-infrared spectroscopy [Yildirim et al., 2018]), Zeleny sedimentation test (AACCI Method 56-60.01 [AACCI, 2010]), delayed Zeleny sedimentation test (Greenaway et al., 1965) and falling number values (AACCI Method 56-81.03 [AACCI, 2010]) were determined. All analyses applied to flour samples were determined two days after flour milling. In the study, tempered flour from each treatment group (single-step 24 h, single-step 48 h, two-step 48 h, two-step 48 h and kept in room conditions for 30 days) was compared with the flour of non-tempered group (control).

Statistical Analysis

All experiments (different tempering treatments applied to wheat samples and analyses) were carried out in two replicates. In the experiments, variance of analysis (ANOVA) was used to obtain conclusion on measured characteristics of wheat and flour samples. When a significant difference was found between the treatments, Duncan's multiple comparison tests was used to determine the difference among treatment means at $P<0.05$. Statistical analysis was performed using the statistical package program developed by the SAS statistical institute (The SAS System for Windows v6.12; SAS Institute, 1982).

RESULTS and DISCUSSION

Basic Characteristics of Wheat Samples

The average values for the physical and chemical properties of two different wheat samples used in the study are presented in Table 1. It has been determined that both wheat varieties are exposed to about 2% sun pest damage ratios. The weight of thousand kernels of both wheat samples was found to be relatively high. Hectoliter weight values indicate that both wheat samples according to Turkish Standards Institution wheat standard (TS 2974; TSE, 2018) are included in the first quality bread wheat class. The yield of Adana-99 wheat variety from wheat samples passed through only two reducing rolls was 52.3% and the flour yield of Russian wheat variety was 61.3%.

Table 1. The average values for the physical and chemical properties of two different wheat samples used in the tudy.
Çizelge 1. Çalışmada kullanılan iki farklı buğday örneğinin fiziksel ve kimyasal özelliklerine ait ortalama değerler.

Wheat Variety (*Buğday Çeşidi*)	Sunn Pest DamageRatio (*Süne Emgi Oranı*)	Thousand KernelWeight (*1000 Tane*	Hectoliter Weight (*Hektolitre Ağırlığı*)	Flour Yield (*Un Verimi*)	VitreousKernel Ratio (*Camsı Tane*	Translucent Kernel Ratio (*Dönme Tane*	Mealy Kernel Ratio (*Unsu Tane*	Moisture Content (*Nem Miktarı*)	Protein Content(*Protein Miktarı*)	Starch Content (*Nişasta Miktarı*)
Adana-99	1.92±0.07	37.0±0.8	83.1±0.6	52.3±0.5	45±3	49±4	6±2	12.8±0.1	13.0±0.1	62±3
Russian	2.15±0.06	40.5±0.8	82.7±0.3	61.3±0.8	15±2	39±3	46±3	14.1±0.1	10.7±0	63±4

[1] Calculation based on dry matter basis.
[2] It was calculated by, milling with using 3 crushing and 2 reducing rolls in CD1 mill and sieving through the mill's own sieve.

The data of the kernel vitreousness analysis revealed that the Adana-99 wheat variety had a relatively hard grain structure whereas the Russian wheat variety had a soft grain structure (Table 1). Data on kernel vitreousness reveals that the characteristics of the two wheat samples discussed in the experiment are different from each other. This situation serves the purpose of planning and implementation of the study. From both wheat samples exposed to the same number of crushing and reducing rolls, as expected, the flour yield of the Russian wheat variety was found to be as high as 17% compared to the Adana-99, which had a hard grain structure. This finding is consistent with the literature findings and reports (Kent, 1984; Delcour and Hoseney, 2010). Namely; more energy is required to obtain a sufficient amount of flour and semolina yield by milling hard wheat. Yet, wheat samples were milled in the same laboratory mill (they were exposed to the same energy consumption) and flour yield of Adana-99 wheat variety was found low.

The weight of hectoliter used in transport and storage of wheat varies depends on wheat type, variety, shape, density, foreign matter content, size and with the characteristics of the spring-winter. The weight of a thousand kernels varies depending on the wheat type and the climate, gives an idea of the size of wheat grains and that they are weak or plump (Uluöz, 1965). In milling, it is desirable to have high data for each of the two measurements. Because the thousand kernel and hectoliter weights are positively correlated with flour yield (Kinaci and Kinaci, 2004; Dizlek and Ozer, 2016a). Although it is a hard and soft feature of the grain depends on wheat variety, it also shows great changes according to growing conditions. Generally, the quality and quantity of gluten in hard-grained wheat is high. The amount of moisture is an important criterion for trade and storage of wheat; depends on climate and storage conditions where wheat grown. The amount of protein in wheat depends on the type, variety, environmental conditions (climate, soil, disease and pests) and production conditions (fertilization quantity and type, irrigation, machine farming) varies between 8-20%. In wheat, the amount and composition of the ash vary depending on the type and varieties, the amount of mineral matter in the soil where the wheat grows, its ability to be taken by the plant, the condition of fertilization and the climate (Kent, 1984).

Moisture content of Adana-99 wheat is below 14%, which can be stored safely (Kent, 1984). The moisture content of Russian wheat is at the critical limit. The initial moisture content of both wheat samples was found to be relatively high. It was thought that it might be effective for the wheat to absorb some moisture from the environment during storage in the cold storage. The amount of protein varying depending on properties of wheat variety and genetic factors, as expected, high in Adana-99% wheat variety (13%), Russian wheat variety was found to be relatively low (10.7%). In relation to the amount of protein, the starch amount of Adana-99 wheat is relatively lower than that of Russian wheat (Table 1). By analyzing the data of Table 1 together; it is concluded that the wheat sample of Adana-99 is medium hard quality wheat, while Russian is relatively soft bread wheat. Both types have superior physical properties; in this sense, thousand kernel, hectoliter weights and flour yields were found to have high values.

Table 2. Some physical and chemical properties of wheat flours milled with different tempering treatments[1].
Çizelge 2. Farklı tavlama muameleleriyle öğütülmüş buğday unlarının bazı fiziksel ve kimyasal özellikleri[1].

Wheat Variety (Buğday *Çeşidi*)	Tempering Treatment (*Tavlama Muamelesi*)	Moisture Content (*Nem Miktarı*) (%)	Ash Content (*Kül Miktarı*) (%)	Protein Content (Protein *Miktarı*) (%)	Damaged Starch (*Zedelenmiş Nişasta*) (%)[2]	Color Properties (*Renk Özellikleri*) L*	a*	b*	Flour Yield (*Un Verimi*) (%)[3]
Adana-99	non-tempered (control)	13.0^{c}±0.1	0.75^{a}±0.02	11.5^{a}±0.3	3.1^{c}±0.1	90.38^{d}±0.2	0.36^{a}±0	8.46±0.5	52.3^{c}±0.3
	single-step tempered for 24 h	5.7^{a}±0.4	0.61^{b}±0.02	10.9^{c}±0	5.6^{a}±0.2	93.43^{a}±0.3	0.14^{d}±0	7.65±0.5	53.6^{b}±0.3
	single-step tempered for 48 h	5.2^{a}±0.3	0.63^{b}±0.02	11.1^{b}±0.1	5.2^{a}±0.2	92.54^{b}±0.2	0.20^{c}±0	7.74±0.6	55.7^{a}±0.7
	two-step tempered for 24 h	5.4^{a}±0.3	0.63^{b}±0.01	11.3^{ab}±0.2	4.4^{b}±0.1	91.49^{c}±0.3	0.29^{b}±0	8.43±0.4	51.9^{c}±0.5
	two-step tempered for 48 h (rested for 30 days)	3.5^{b}±0.1	0.63^{b}±0.02	10.9^{c}±0.1	4.3^{b}±0.1	92.77^{ab}±0.2	0.17^{cd}±0	8.06±0.6	
Russian	non-tempered (control)	4.2^{b}±0.2	0.59^{a}±0.01	8.8^{ab}±0.2	4.5^{d}±0	93.04^{c}±0	0.12^{a}±0	7.93±0.5	61.3^{c}±0.3
	single-step tempered for 24 h	5.0^{a}±0.1	0.56^{b}±0	8.6^{b}±0	5.4^{b}±0.1	94.36^{a}±0.3	0.08^{b}±0	7.18±0.4	60.8^{c}±0.5
	single-step tempered for 48 h	5.2^{a}±0.3	0.54^{c}±0	8.8^{ab}±0.2	6.0^{a}±0.1	94.14^{a}±0.3	0.08^{b}±0	7.61±0.3	63.4^{a}±0.2
	two-step tempered for 24 h	5.5^{a}±0.5	0.52^{d}±0.01	9.0^{a}±0.2	4.8^{c}±0.1	93.19^{b}±0.1	0.13^{a}±0	8.15±0.5	62.8^{b}±0.1
	two-step tempered for 48 h (rested for 30 days)	3.6^{c}±0.1	0.52^{d}±0	8.5^{b}±0.1	4.8^{c}±0.1	94.35^{a}±0.2	0.06^{c}±0	7.72±0.5	

[1] Mean values in the same column and same wheat varieties (Adana-99 or Russian) followed by different superscript letters are significantly different ($P<0.05$).
[2] It is given on the dry matter basis.
[3] It was calculated by, milling with using 3 crushing and 2 reducing rolls in CD1 mill and sieving through the mill's own sieve.

Effects of Different Tempering Treatments on Physical and Chemical Properties of Flour

Some physical and chemical properties of wheat flour milled with different tempering treatments are given in Table 2. Applying tempering process to both wheat samples and followed milling, moisture content of flour was found higher than non-tempered control sample. In the case of relatively high (15% to 15.7%) moisture values of flour samples; the fact that the flour was not rested and the moisture analysis was carried out on the second day following the milling were effective. It was determined that the moisture content of the wheats was reduced to a reasonable rate (13.5%-13.6%) after the flour of the two-step tempered wheats were rested for 30 days.

As expected, the amount of ash obtained from wheat samples decreased with the application of tempering (Table 2). In the Adana-99 wheat, different number (1 and 2) and time (24 and 48 h) the tempering process did not cause a change in the amount of ash in itself, whereas in Russian wheat sample the amount of ash samples decreased in parallel with the increase in the number and duration of the tempering ($P<0.05$). The finding obtained from the study of the tempering process which is easier to break the wheat by loosening the rigid bond between the shell and the endosperm layers of wheat and to obtain the high yield flour with low-ash color values is consistent with the literature finding (Ibanoglu, 2001). It was determined that the data of the ash analysis were consistent with the findings of the physical analysis of the samples. Accordingly, the ash content of Adana-99 wheat flour, which has a hard grain structure and low flour yield, is significantly higher than Russian wheat flour.

As expected in the Adana-99 sample, the amount of protein of non-tempered wheat samples was higher than that of tempered wheat samples. This indicates that the bran contamination of the non-tempered wheat is more. Different time and number of tempering treatments resulted in easier separation of the endosperm from the crust and thus decreased protein content of the flour samples. As a matter of fact, the shell (bran) layer of wheat contains higher rates of protein than the endosperm (flour) layer (Kent, 1984; Delcour and Hoseney, 2010). There was no meaningful relationship between protein values of flour of Russian wheat samples. More limited variation was observed between the protein amounts of the tempering treatments of the Russian variety according to the Adana-99 sample (Table 2).

In both wheat samples, it was determined that the tempering treatment increased the amount of damaged starch compared to the control sample. It has been found that once made tempering has increased starch damage in both Adana-99 and Russian wheat more than the tempering process performed twice.

As expected, the tempering process was found to cause bleaching of flour color (Table 2). In both wheat flour samples, it was determined that the control sample had a darker color than the tempered samples. It was found that there was a significant relation between L^* values and ash amount of flour samples. Flour samples that were rested for 30 days were found to be lighter color compared to untreated flour samples. In this situation, it is thought that resting with oxidation event is effective in natural bleaching of flour. The negative correlation was found between the amount of ash obtained from the study and the L^* value, and a positive correlation was found with a^* value. It was observed that the data were consistent with the findings of Alfin and Çakmaklı (1999). Reduction in redness of flour samples was observed with the treatment of tempering. It has been found that this situation is consistent with the amount of ash as explained above. However, the redness value of the flours of wheat samples, two-step tempered for 48 h, increased. This increase led to the same red value as the control sample of the Russian wheat variety obtained by the said treatment. In both wheat flour samples; a meaningful relationship was not found between b^* color values and tempering treatments. However, in general, the control samples were observed to have a more yellowish color than the other three samples except sample with two-step tempered for 48 h. By examining the effect of different tempering treatments on the amount of flour obtained from wheat samples, it was determined that the samples which were tempered once for 48 h in both varieties had the highest flour yield. The tempering process did not affect flour yield in Adana-99 variety but increased the total flour yield by 1.5% in Russian variety. Investigation of flour yield findings together: the tempering process increases the flour yield of the wheat samples as can be expected, the two-step tempering process does not show the expected benefit at the point of increasing efficiency, once the tempering process is better, but it is more effective for 48 h - both wheat varieties. Hook et al. (1982); who study on the subject, reported that an increase in the amount of tempering water in wheat grain reduced the yield of flour. Therefore, the researchers stated that the tempering process should be balanced in such a way that the flour yield and the ratio of bran in the flour are acceptable. The data obtained from this study were partially conducted by Hook et al. (1982). This situation is thought to be caused by the different types of wheat used in the studies and the amount of water given in tempering process.

Effects of Different Tempering Treatments on the Technological Properties of Flour Physicochemical properties of flour samples are presented in Table 3. In Adana-99 sample, the wet gluten amount of non-tempered wheat flour was found to be higher than the tempered treatments. This situation is thought to be caused by easier treatment of crust-endosperm decomposition and easier obtaining of pure endosperm flour. The amount of the gluten proteins increases from the center outward in the endosperm (Delcour and Hoseney, 2010). For this purpose, a relatively high concentration of aleurone layer and endosperm outer layer flours were found to be relatively high in non-tempered wheat flour samples. It has been found that the wet gluten of wheat flour sample with two-step tempered for 48 h is relatively higher than the other tempered treatments. However, it was found that there was no significant difference (P>0.05) between the tempering treatments in terms of wet gluten content. In the Russian sample, it was observed that there was no statistical difference in wet gluten amount between five different treatments. Generally, dry gluten values were found to be compatible with wet gluten values and it was determined that there was a limited variation in dry gluten content between tempering treatments. The quantity and quality of the gluten; is one of the most important quality criteria for wheat and it is one of the most important factors affecting the dough: kneading, processing properties, gas holding capacity and final product quality (Park et al., 2006; Delcour and Hoseney, 2010; Dizlek and Ozer, 2016b; 2017; Girard et al., 2019). It is desirable for a bread flour to have high gluten quantity and quality (Dizlek and Ozer, 2016 a; b).

Table 3. Physicochemical properties of wheat flours milled with different tempering treatments[1].
Çizelge 3. *Farklı tavlama muameleleriyle öğütülmüş buğday unlarının fizikokimyasal özellikleri[1].*

Wheat Variety (*Buğday Çeşidi*)	Tempering Treatment (*Tavlama Muamelesi*)	Wet Gluten Content (*Yaş Gluten Miktarı*) (%)	Dry Gluten Content (*Kuru Gluten Miktarı*) (%)	Sedimentation Value (*Sedimantasyon Değeri*) (ml)	Delayed Sedimentation Value (Gecikmeli *Sedimantasyon Değeri*) (ml)	Falling Number Value (*Düşme Sayısı Değeri*) (s)
Adana-99	non-tempered (control)	$28.5^{a}\pm0.4$	$9.2^{a}\pm0.1$	$38^{a}\pm1$	$31^{c}\pm0$	337±21
	single step tempered for 24 h	$27.0^{b}\pm0.2$	$8.9^{b}\pm0$	$36^{ab}\pm1$	$41^{a}\pm1$	344±18
	single step tempered for 48 h	$27.3^{b}\pm0.2$	$9.0^{ab}\pm0.1$	$38^{a}\pm1$	$38^{a}\pm2$	344±21
	two-step tempered for 24 h	$27.9^{ab}\pm0.3$	$9.1^{a}\pm0.1$	$34^{b}\pm1$	$39^{a}\pm2$	349±19
	two-step tempered for 48 h (rested for 30 days)	$27.3^{b}\pm0.2$	$8.8^{b}\pm0.1$	$30^{c}\pm1$	$34^{b}\pm1$	362±20
Russian	non-tempered (control)	21.6±0.4	$6.7^{b}\pm0.1$	$32^{a}\pm1$	$20^{b}\pm1$	356±24
	single step tempered for 24 h	21.5±0.5	$6.8^{ab}\pm0$	$30^{a}\pm1$	$33^{a}\pm2$	351±19
	single step tempered for 48 h	21.5±0.3	$6.9^{a}\pm0.1$	$27^{b}\pm0$	$34^{a}\pm2$	363±19
	two-step tempered for 24 h	22.0±0.4	$7.0^{a}\pm0.1$	$31^{a}\pm1$	$32^{a}\pm3$	378±21
	two-step tempered for 48 h (rested for 30 days)	21.3±0.4	$6.6^{b}\pm0.1$	$25^{c}\pm1$	$33^{a}\pm3$	364±18

[1] Mean values in the same column and same wheat varieties (Adana-99 or Russian) followed by different superscript letters are significantly different (P<0.05).

It is reported that there is correlation between sedimentation values, which is one of the most important criteria used in determining the quality of wheat and vary depending on protein (gluten) quality, and results of baking experiments (Faridi and Faubion, 1990; Dıraman, 2010). By examining the findings of the sedimentation value which is a very important quality criterion in flour milling and showing the gluten quantity-quality simultaneously, both wheat varieties had lower sedimentation value compared to the control sample and there was some decrease in gluten quantity and quality. Generally, the findings obtained from sedimentation value were found to be compatible with wet-dry gluten content values. It was observed that the flour obtained by milling the samples with tempered two-step for 48 h and rest 30 days in both wheat varieties had the lowest sedimentation value. According to the data of delayed sedimentation analysis, the tempering process results in improvement in gluten quantity and quality of the samples as expected, there is no significant difference (P>0.05) between the different tempering times and numbers in terms of these improvements, but according to the control sample, both tempering for 24-48 h and tempering for 1-2 times showed a significant effect (P<0.05) on the gluten quality of the samples. This is due to the triggering of protease activity by the 30 °C temperature and 2 h time norms applied in the delayed sedimentation analysis in the control sample which has sunn pest damage but is not subjected to any tempering treatment. With the tempering process, gluten quality is improved (gluten structure firming) and hence the delayed sedimentation values of the flour of tempered samples are increased (Ertugay et al., 1995c; Dıraman et al., 2013; Dizlek and Islamoglu, 2015).

By analyzing the falling number values that provide information about the amylase activity of flour samples (Table 3), it was determined that there was no significant difference (P>0.05) between the tempering treatments applied in both wheat samples. This finding is consistent with the statement of Sünter (2003); which investigated the effect of different tempering times and temperatures on wheat quality. The data on falling number analysis shows that it would be beneficial to add a certain (relatively less) amount of α-amylase preparation and / or malt flour to the dough formulation in the bread making with of both wheat samples flour.

In literature, there is a little information about two-step tempering. According to Buhler handbook (Buhler 2016); general rules for tempering in two-steps as follows: first tempering: 2/3 of total tempering time (water addition to about 1-2% lower than target moisture) and second tempering: 1/3 of total tempering time (it should be 8 – maximum 12 h, no longer leads to drying out of bran; water addition 1-2%). In this study, two-step tempering treatment was partially effective on the physical, chemical and technological properties of flour in bread wheat (*Triticum aestivum* L.). However, further studies are needed to elucidate on two-step tempering process.

CONCLUSIONS and RECOMMENDATIONS

In this study, the effects of two-step tempering treatment on the physical, chemical and physicochemical characteristics of two different bread wheats were investigated. In this sense, the effects of two different tempering times (24 and 48 h) and two different tempering applications (once or twice) on the raw material (wheat) and intermediate product (flour) were investigated. In addition, the wheat flour with two-step tempered, for the purpose of setting a precedent for the market conditions, has been evaluated as analytically by resting for 30 days. The findings obtained from the study are summarized below:

The tempering treatment resulted in a limited improvement in the physical (flour yield and color), chemical and technological properties of the flour. It was found that, especially more evident in the ash content, the tempering treatment reduced the ash and protein contents of the flour samples, however it resulted in an increment in the gluten quality (The tempering treatment resulted in a significant increase in the value of delayed sedimentation according to the normal sedimentation value of flour samples). Tempering treatment for 48 h among single-step tempering treatments resulted in more improved flour quality. As a result, the findings include clues that, water when added to the wheat in the tempering process in two-steps results in an improved flour quality. However, further studies are required on this subject to elucidate the mechanism.

It is recommended that in subsequent studies, the kinetics of two-step tempering treatment applied to wheat and its effect on product properties can be examined more comprehensively. If this and similar studies are carried out on more wheat varieties, more inclusive analytical findings can be achieved. The study was carried out by applying the cold tempering method. In the subsequent studies, the effect of two-step tempering on wheat, flour, dough, and bread quality can be investigated by use of warm and / or hot tempering methods. It is predicted that these practices will further improve the bread wheat quality. In addition, this study can be continued with a different research in terms of the energy spent in a pilot flour mill.

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Author's Contributions

All authors contributed to the study conception and design. Mustafa KURT has provided research materials, milled the wheat groups and performed the laboratory work. Halef DIZLEK performed the statistical analysis, interpreted the results and drafted the manuscript. The first draft of the manuscript was written by Halef DIZLEK. Tempering process of wheat samples and writing - original draft preparation of the manuscript were written by Mustafa KURT and Halef DIZLEK; Writing - review and editing were performed by Halef DIZLEK. All authors commented on previous versions of the manuscript. Also all authors read and approved the final manuscript. Halef DIZLEK is the Master of Science supervisor of Mustafa KURT.

Statement of Conflict of Interest

Authors have declared no conflict of interest.

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