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Determination of the effect of high energy ultrasound application in tempering on flour quality of wheat



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ABSTRACT

This research has been carried out under laboratory conditions. To determine the effect of soaking with ultrasound application to dampen hard wheat samples at one stage in tempering process was the prior goal. In the experiments, Bezostaya-1 wheat samples in different hardness levels (45, 65 and 75%) are used as material. The milling quality and qualitative properties of the flour were investigated. Results compared with the classic tempering process which has two stages tempering for hard and semi-hard wheat at the industrial applications. Ultrasonication of the samples has been applied by a probe-type ultrasound device. Because of experimental deviations, inevitable positive effect of ultrasonic tempering had not seen in the quality parameters of milling experiments like flour yield and energy consumption. But tempering with ultrasound application increased the speed of water intake and diffusion into the grain center. In the study, the wet gluten rates of the flour obtained by classical tempering process were 24.52% and the gluten index rate was 87.17%, while the results obtained by ultrasonic tempering were 22.70% and 93.33%, respectively. Thus it raised flour quality a little, possibly, due to low amount but better quality gluten coming from central endosperm in the flour obtained. Other analysis results in flour showed significant differences in some values of classical and ultrasonic tempering processes. As a result of ultrasonic tempering, the fineness rate (granulation) increased from 73.27% to 79.77%, ash content decreased from 0.61% to 0.55%, Zeleny sedimentation value decreased from 25.0 mL to 23.67 mL, flour stability increased from 9.76 min to 12.06 min, water absorption 59.1% decreased from 61.28%, softening resistance increased from 400.33 BU to 504.50 BU, maximum resistance increased from 420.50 BU to 536.16 BU. In bread trials, bread volume increased from 328.3 mL to 347.3 mL, and specific volume increased from 2.39 mL / g to 2.57 mL / g. These issues have been confirmed by some analysis such as fine granulation, low ash content, high gluten index, some farinogram, and extensigram properties, and finally better performance in bread making was observed.

1. Introduction

Considering today's needs, qualitative specifications of wheat and flour have great importance in providing quality of the final product [1-8]. Therefore, the selection of raw materials with its quality factors and milling technology applied to be looked for in flour specification. One of the important steps in milling operations is tempering which helps the separation of endosperm from the bran layers and to make easy grinding [9–11].

Tempering operation provides an increment in grinding quality and makes the physical structure of wheat kernel more suitable for grinding. Thus, optimum water level is supplied to kernel and it is rested for a while in tempering silos. The water given to wheat kernel during tempering operations, enters in, and diffused into the center of kernel during resting time [12,13,14]. Tempering is one of the important operations which decrease energy consumption while increase flour yield and especially flour quality in the flour milling industry [15]. There are some different factors, affecting tempering quality positively in the milling technologies. The most important ones of them are the quantity and temperature of tempering water and also the tempering time and mechanical factors like mixing, vibration and scraping [9,16–18].

Ultrasound operation is widely used in the food industry [19–41]. In grain operation, there is a highly limited application area. It can be used in drying and sieving operations [33,41–54]. It is used in malt production to decrease the soaking process and germination time [55–59]. The use of low energy in MHz range, high-frequency ultrasound and high energy, low frequency in kHz range has been subject to research and development for many years [60–67]. There are literature on the

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fact that the ultrasound technique is used to affect enzymatic activity, to increase yield in the production of corn rice starch and to shorten steeping time steeping of legumes with the effect of cavitation [39,68–75]. There has been no source on the use of ultrasound applications in tempering process of flour milling.

The effect of high and low energy sonication operation on protein structure of wheat was examined by Singh and McRat [76] using probetype ultrasound equipment. Low energy sonication application increased gluten in sulfhydryl content in solution. There was a decrease in sulfhydryl groups in gluten solution during high energy sonication applications. High energy sonication led to the formation of new disulfide bonds with an association of free cysteine molecules in gluten protein.

In this research, it is estimated that the soaking operation with an ultrasound application can be effective in tempering operation in flour milling, as it leads to an increase in mass transfer [37,77–80]. and damage to anatomic layers of wheat kernel, increasing the endosperm and bran separation in milling effectively. Therefore, it is aimed to shorten tempering time, increase the effect of tempering operation, decrease investment, maintenance and workmanship expenses in flour milling mechanization and increase in milling quality.

2. Materials and methods

2.1. Materials

In this study, the Bezostaya-1 wheat sample at different hardness levels is used as materials, which were received from Afyonkarahisar market. At the preparation of the samples, the material cleaned from foreign material and homogenized. Sieves, having diameters between 2.25 and 3.5 mm, are used to remove the extreme sized kernels. To obtain different hardness degrees at the samples, the material was calibrated according to the Grobecker section test, to 45, 65 and 75% kernel hardness levels. The chemical and physical properties of wheat of different hardness used in the study are given in Table 1.

In the tempering process, the water at medium hardness (120 ppm) was used for dampening. In ultrasound application, prob type GEX-600 Ultrasonic processor equipment, working at 20 kHz frequency and 600 W power was used. Trials were conducted in two steps. In the first step, analytic quality specifications of material were detected (Table 1) and with pretesting the optimization of operational parameters was supplied; in second steps, the studies on the effects of ultrasonication on the quality parameters were obtained. All tempering operations made away by using optimum operational parameters (100% amplitude, one minute soaking time, 24 h rest time). In this context, firstly, milling properties were determined according to the experimental design of "three kernel hardness (45, 65, and 75%), two tempering methods (classic and ultrasound applied" for two replication according to the factorial plan of "(3x2)x2". And the all sample combinations obtained

Table 1

Chemical and physical properties of wheat of different hardness.

were conditioned for 24 h and milled at standard conditions. Then the flour samples obtained were rested for 3 weeks for maturation, afterward their physical, chemical and technological specifications were examined.

2.2. Methods

Kernel size and homogeneity, hectoliter weight (kg/hL) thousand kernel weight (g) and hardness (%) determinations were done according to Pomeranz Y (1988) and Elgün et al (2005) for three wheat samples, used as materials [14.81.82]. Color values of wheat samples. flours, and bread were measured as L^* value [(0) black to (1 0 0) white]. a^* value [(+) red, (-) green] and b^* value [(+) yellow to (-) blue], using Konika-Minolta CR-400 (Kornica Minolta Sensingg Inc, Osaka, Japan), in *L**, *a** and *b** values [83]. Moisture, protein and ash quantity determinations were done for wheat and flour samples. The water amount was calculated with AACC 44-19 method. Protein determination of these samples was conducted by the Kjeldahl method (AACC 46-12), and ash determination was performed according to ICC Standard No. 104/1 method [84]. The results were given based on dry matter. Zeleny sedimentation was conducted according to AACC Method 56-60, gluten determination was conducted according to AACC 38-12 [85], falling number value was conducted according to AACC 56-816 [86]. Tempering of the samples was applied to manually cleaned wheat samples with dampening and resting operations under laboratory conditions. In dampening, the soaking method was conducted by waiting for wheat samples in water for a certain time. Two different soaking methods as "ultrasonic method" and "classic method" without US application were used. Within this context, wet gluten amount, gluten index, Zeleny sedimentation test, and falling number were determined [84,85,87,88]. For wet gluten amount and index test (AACC 38-12) determinations. Glutomatic-2200 wash machine and centrifuge system devices were used [89]. Farinogram specifications of flour samples, obtained according to experimental designs, were determined according to ICC Standard Method No 115/1, extensigram specification of them were determined according to ICC Standard Method No: 114/1 [84]. Bread was made by modifying the method given by AACC 10–10 [85]. The ingredients were kneaded until mature dough was obtained (Hobart N50, Canada) and rested at 30 °C and 80-90% relative humidity for 50 min and baked at 230 °C for 25 min (Arcelik, ARMD 580, Turkey). After the loaves come out of the oven and cool down, their weights and volumes were measured and after an hour they were put into polyethylene bags and sealed. For other bread specifications, sealed bread in polyethylene bags was held for 24 h at room temperature. Color measurement of bread crust and the crumb was done by Hunter colorimeter, measuring L^* , a^* and b^* values [83].

Kernel Hardness (%)	Some Cher	nical proper	ties					
	Moisture (%)	Ash (%)	Protein (%) ³	Zeleny sed	imentation (mL)	Wet gluten (%)	Falling Number (s)
45	9.6		1.77	11.3	20		20.0	341
65	10.3		1.79	11.9	25		22.9	377
75	9.4		1.84	13.1	30		30.6	380
Kernel Hardness (%)	Some Phys Color	e Physical properties r Hectoliter wei		Hectoliter weig	ght (kg/hL)	Thousand kernel	s weight (g)	Kernel homogeneity (%)
	L^*	a*	b^*					
45	50.7	7.9	19.9	80.2		38.7		97.85
65	50.5	8.5	19.7	80.67		36.74		97.3
75	47.9	7.5	18.0	80.83		35.83		98.3

2.3. Statistical analyses

The data obtained during the experiments were subjected to variance analysis and the mean of statistically important main variation sources was compared with Duncan's multiple Range Test. The results of statistical analysis were summarized in tables; statistically important interactions were discussed on figures [90]. TARIST test assessment packet (Version 4.0) computer program was used for statistical analyses.

3. Experimental results

3.1. The effect of the time of soaking with ultrasound application in tempering on the milling and flour quality of wheat

The data obtained in the research subjected to analysis of variance (ANOVA) and the results important at a statistically significant level (P < 0.05) evaluated and discussed below:

3.1.1. Flour yield

The results of the analysis of variance (ANOVA) showed that "kernel hardness" and "soaking time" were significantly effective on flour yield (Table 2). While increasing kernel hardness increased the flour yield as expected, but increasing soaking time decreased the yield due to excessive water derived from longer application times. Ultrasound (US) application stayed ineffective on flour yield because of experimental deviations as a result of the milling studies with small samples on lab mill and the significant two-way interactions at P < 0.01 level. Possibly, the expected difference in flour yields due to effect of US application was covered by the "kernel hardness × tempering method" and "tempering method × soaking time" interactions. In both tempering methods, 30 s soaking time with US application for the all hardness level was the most suitable time to get optimum water absorption needed for tempering.

3.1.2. Flour fineness

As seen in Table 2, under 140- μ m sub sieve material of the flour samples was not significantly affected (P < 0.05) by all main variation sources. There is an increment in flour fineness together with soaking interval a little bit but not significantly (P < 0.05). Descriptively, there is a meaningful difference between both tempering methods. Ultrasonic tempering (87.30%) showed finer flour granulation than that of classic one (84.77) as seen in Table 2.

3.1.3. Flour moisture

Flour moisture is an important factor not only for the water immigration into the kernel but also at the immediate flour yield just after the milling operations as milling yield. As a result of ANOVA, flour moisture content was not influenced by the US tempering method significantly (P < 0.05). The optimum moisture needed for tempering at all hardness grades, including 75%, was provided for 30 s. The higher soaking time will cause high flour moisture, but low flour yield and ash amount. It shouldn't be ignored that high tempering water decreases flour yield together with ash amount (Table 2) [17].

3.1.4. Flour ash

In flour milling, flour ash amount is a very important parameter for milling quality [91]. The results of ANOVA showed that all factors treated were not significantly effective on flour ash level (P < 0.05). As a result, there is no difference between the two methods in flour ash amount indicating the same tempering, flour and milling quality. Possibly, the US tempering method with 30 s soaking time could give a flour with lower ash than that of classic one (Table 2).

3.1.5. Wet gluten amount and gluten index

In flour milling and baking science, flour wet gluten amount and gluten index are very important parameters in the estimation of wheat and flour quality [46]. The results of ANOVA showed that all factors treated were not effective on both quality parameters (P < 0.05). As a result, there is no difference between the two methods in flour quality indicating at least the same tempering, flour milling performance and baking quality. As a result of the descriptive assessment, possibly the US tempering method with 30 s soaking time could give a flour with a little bit lower wet gluten but with a higher gluten index than those of classic ones (Table 2).

These results showed that hard wheat kernel samples could take up the water enough in 30 s soaking interval at only one stage in dampening and also not different than those of classic tempering method with two stages. There is no difference between two methods in milling performance as seen in the yield, water amount and granule size of flour. Possibly US tempering method gave a flour with finer granulation than the classic one (Table 2).

3.2. The effect of soaking with ultrasound application in the tempering of wheat on milling and flour quality

These data about the milling quality of the samples were obtained with optimum US tempering conditions with 30 s soaking time. According to the results of ANOVA, the significant main variation sources subjected to Duncan's multiple range test (P < 0.05). The results are summarized in Table 3.

3.3. The effect of the soaking with ultrasound application in tempering of wheat on milling quality

3.3.1. Flour yield

According to ANOVA results, it is determined that kernel hardness and tempering methods have no significant effect on flour yield

Table 2

The Changes of	Oualitative Pro	perties of T	The Flour Dar	npened Different	Soaking	Times by	Classic and	Ultrasonic 7	Fempering	Methods ¹
	· · · · · · · · · · · · · · · · · · ·									

Variation	Flour Yield	Granule Fineness ²	Flour Moisture	Flour Ash ³	Wet Gluten	Gluten index
Sources Kernel Hardness	(%)	(%)	(%)	(%)	(%)	(%)
45%	$68.960^{b} \pm 0.58$	$86.117^{a} \pm 2.8$	$15.867^{ab} \pm 0.37$	$0.569^{a} \pm 0.01$	$24.717^{b} \pm 0.6$	$92.917^{a} \pm 3.6$
65%	$69.105^{b} \pm 0,95$	$86.225^{a} \pm 2,4$	$16.125^{a} \pm 0,33$	$0.580^{a} \pm 0.01$	$25.650^{\rm b} \pm 1.5$	$92.667^{a} \pm 3.9$
75%	$70.280^{a} \pm 0.82$	$85.767^{a} \pm 2,5$	$15.708^{b} \pm 0,38$	$0.573^{a} \pm 0,01$	$33.650^{a} \pm 1,1$	$83.417^{b} \pm 3.8$
TemperingMethod						
Classic	$69.406^{a} \pm 1.1$	$84.772^{a} \pm 2,2$	$15.789^{\rm a} \pm 0,4$	$0.577^{a} \pm 0,01$	$28.228^{a} \pm 4,2$	87.333 ^b ± 5,9
Ultrasonic	$69.491^{a} \pm 0.8$	$87.300^{a} \pm 2.3$	$16.011^{a} \pm 0.3$	$0.571^{a} \pm 0,01$	$27.783^{a} \pm 4,3$	$92.000^{a} \pm 4,7$
Soakig Time						
30 s	$69.898^{a} \pm 1,2$	$85.425^{a} \pm 2.4$	$15.683^{b} \pm 0.3$	$0.569^{a} \pm 0,01$	$28.083^{a} \pm 4,5$	$88.250^{a} \pm 5,5$
45 s	$69.214^{b} \pm 0.8$	$86.425^{a} \pm 3.1$	$15.883^{ab} \pm 0,3$	$0.577^{a} \pm 0,01$	$28.433^{a} \pm 4,5$	$90.083^{a} \pm 6,0$
60 s	$69.233^{b} \pm 0,6$	$86.258^{a} \pm 2,0$	$16.133^{a} \pm 0,2$	$0.576^{a} \pm 0,01$	$27.500^{a} \pm 3.8$	$90.667^{a} \pm 6,0$

 1 The means with the same letter are not different than the others (P < 0.05). $^{2:}$ Under 140 μ . 3 Based on dry matter

	Flour								Wet	Gluten	Zeleny	Falling
Variation	Yield	Fineness ²	Color			Moisture	Ash ³	Protein ⁴	gluten	index	Sedim.	number
Sources	(%)	(%)	L^*	a*	p^*	(%)	(%)	(%)	(%)	(%)	(mL)	(s)
Kernel Hardness												
(%)												
45	$68.15^{a} \pm 0.75$	$76.83^{ab} \pm 2,39$	$95.52^{a} \pm 0.56$	$-0.22^{a} \pm 0,10$	$8.95^{a} \pm 0,20$	$16.52^{a} \pm 0,46$	$0.593^{a} \pm 0.04$	$8.30^{c} \pm 0,26$	$19.68^{b} \pm 0.67$	$93.00^{a} \pm 5,59$	$19.50^{c} \pm 0.57$	$346.25^{b} \pm 8,34$
65	$68.28^{a} \pm 0,50$	$71.68^{b} \pm 11,27$	$95.50^{a} \pm 0,31$	$-0.27^{\mathrm{a}} \pm 0.17$	$8.97^{a} \pm 0.55$	$16.56^{a} \pm 0.68$	$0.588^{a} \pm 0.01$	$8.86^{b} \pm 0,40$	$20.43^{b} \pm 2.94$	$94.75^{a} \pm 5,85$	$23.50^{b} \pm 1,73$	$376.25^{a} \pm 2,21$
75	$67.88^{a} \pm 0.63$	$81.05^{a} \pm 1,00$	$94.34^{b} \pm 0,69$	$-0.20^{a} \pm 0.07$	$8.64^{b} \pm 0.14$	$15.64^{b} \pm 0,32$	$0.565^{a} \pm 0.05$	$11.80^{a} \pm 0,30$	$30.73^{a} \pm 0,25$	$83.00^{b} \pm 3,36$	$30.00^{a} \pm 0.00$	$381.50^{a} \pm 5,19$
Tempering Method												
Classic	$67.82^{a} \pm 0.50$	$73.27^{a} \pm 9,36$	$94.79^{a} \pm 0.63$	$-0.27^{\mathrm{a}}\pm0.12$	$9.02^{a} \pm 0.40$	$15.88^{a} \pm 0.38$	$0.613^{a} \pm 0.01$	$9.74^{a} \pm 1.76$	$24.52^{a} \pm 4,90$	$87.17^{a} \pm 6,96$	$25.00^{a} \pm 4,47$	$365.67^{a} \pm 19,61$
Ultrasonic	$68.39^{a} \pm 0.59$	$79.77^{a} \pm 1.58$	$95.45^{a} \pm 0.77$	$-0.18^{a} \pm 0.10$	$8.69^{b} \pm 0.19$	$16.59^{a} \pm 0,67$	$0.550^{b} \pm 0.02$	$9.57^{a} \pm 1,64$	$22.70^{b} \pm 6,36$	$93.33^{a} \pm 6,25$	$23.67^{b} \pm 5,08$	$370.33^{a} \pm 15,55$

Table 3

¹ The means with the same letter are not significantly different (P < 0.05), ² flour under 140µ sieve, ³³Based on dry matter, ⁴Based on dry matter and Protein: with N*5.7



Fig. 1. The effect of ultrasound application in tempering on granule fineness (< 140 μ) of flour as a result of "Kernel hardness \times Tempering method" interaction in granule fineness.

statistically (Table 3), just as the first study performed before (Table 2). In addition, the ultrasonic tempering method (68.38%) gave a little more flour yield than that of ultrasonic one (67.81%). It is expected that at industrial conditions this little difference may cause a reasonable increase in flour yield.

3.3.2. Flour granule fineness

Fine granulation without starch damage of wheat flour is a good parameter that has been used to estimation of milling quality [92,93]. ANOVA results show the significant effect of the all variation sources at P < 0.05 significance. Averages of granular fineness values of the flours were compared by Duncan's multiple range test. As seen in Table 2, kernel hardness and tempering method caused to change in the granule size of flour. Granule fineness increased together with the US application in soaking. As shown in Fig. 1, according to the "Kernel hardness \times Tempering method" interaction there is a positive effect of US application in tempering process with 30 s soaking. It was more effective for 45 and 65% hardness levels but not for 75%. Possibly this favorable effect is covered by the deficient tempering water or US application time used for soaking for the sample with 75% hardness.

The soaking with ultrasound application speeds up water immigration into kernel and pixelization of the endosperm was accelerated. Thus, the fineness of flour was increased by US tempering and possibly, fine granulation caused to light color appearance.

3.3.3. Flour color

The results of ANOVA showed some good results about the effect of US application in the tempering process (P < 0.05). According to Duncan's multiple range test results (Table 3), As kernel hardness increased from 45 to 65%, L^* and b^* values did not change significantly. However, at 75% kernel hardness L^* and b^* values decreased significantly. Redness of the flour was not significanly affected by the kernel hardness. The decreasing trend of the redness and yellowness with the increasing of kernel hardness to 75% could cause the increasing of flour whiteness. (P < 0.05). Thus US application in soaking for 30 s gave the flour with more bright and white than those of classic methods (Table 3).

According to the results of ANOVA, "kernel hardness \times tempering method" interaction was found statistically significant (P < 0.01) on yellow color intensity (b* value) of flour. In the going of this interaction given in Fig. 2., 30 min soaking time with US application in the



Fig. 2. The going of "Kernel hardness \times Tempering method" interaction in b^{\star} values.

tempering process gave the best physical structure to the kernel with 65% hardness for the milling operation. Presumably, while the water amount is more than needed to temper the kernel with 45% hardness, it is not enough to the kernel with 75% hardness. These findings showed to be a relationship between kernel hardness and soaking time with US application in tempering like the same as that in classical tempering. It is well-known that kernel hardness and resting time interval is more effective factors in the amount of water given and the resting time interval of the tempering process.

Together with a more effective tempering process, finer, whiter and brighter flour was obtained. There is literature information about the fact that granular and coarse-grained flours have a yellow color appearance, in contrast to fine granulated ones have white and bright color [14,17,92,94,95].

3.4. The effect of ultrasound application in dampening of wheat on some chemical properties of flour

These data about the flour qualities were obtained with optimum US tempering conditions with 30 s soaking time. According to the results of ANOVA, the significant main variation sources subjected to Duncan's comparison test (P < 0.05) and the results are summarized in Table 3.

3.4.1. Moisture amount

According to ANOVA results, it is determined that both "Kernel Hardness" and "Tempering Method" have statistically significant effect (P < 0.01) on moisture amount of flour after three weeks rest following milling process. In the results of Duncan's multiple range test (Table 3), it was seen that the moisture amount of flour suddenly fell in flour at 75% kernel hardness. This data may be proof of the fact that the water can't be absorbed enough and not immigrate into central endosperm within 24 h rest time in the tempering process. This event is also a possible cause of the poor temper of samples with 75% kernel hardness as being discussed before in milling quality. The effect of tempering method on the moisture amount of flour was shown diagrammatically in Fig. 4. It shows that the ultrasonic tempering method caused more water immigration into and to save water in endosperm also in flour versus to that of classic one. This situation shows that the water reached to central endosperm up to thin cell walls was kept inside the kernel easily. Also, the loss of water by evaporation from outer layers decreases during the rest time of the tempering process [14,16,17,93]. As a result, US application in dampening possibly will cause to better events than those of classic one in water absorption and immigration into kernel and in the tempering of wheat, not only in time



Fig. 3. The going of "Kernel hardness \times Tempering method" interaction in wet gluten.



Fig. 4. The going of "Kernel Hardness \times Tempering Method" Interaction in development time.

and investment saving but also an increase in milling and flour quality even though with higher flour moisture after three weeks aging time (Table 3).

3.4.2. Ash amount

Ash amount of flour plays an important role to achieve well-raised bread. In bread obtained from flour with high ash content, the volume increase is not at the desired level. [96,97]. As the amount of ash in the flour increases, the ability to hold gas in the bread dough decreases. [96,98]. The ANOVA results showed that only "Tempering method" has statistically significant effect (P < 0.01) on ash amount of flour. The results of the Duncan test are shown in Table 3. While the average ash amount, obtained by classical tempering was 0.61%, it was decreased to 0.55% by the tempering with ultrasound application. According to this result, it could be stated that ultrasound application in the soaking phase of tempering could positively affect the tempering process by increasing temper effect and flour fineness together with its parity. As a result, at stable milling conditions, US tempering method gave better results than those of classic one, while decreasing the ash amount of

flour, increased the flour fineness. This phenomenon can be considered as a measure of increasing flour yield and milling efficiency for the same ash level in industrial conditions [17,93].

3.4.3. Protein amount

ANOVA results showed that the tempering method did not affect the protein amount of flour. When it is descriptively considered, there is a little decrease in the protein amount obtained by tempering with ultrasound application (9.573%) versus that of classic one (9.738%) (Table 3). According to ANOVA results, the effect of "Kernel Hardness" on protein amount was considered as statistically significant (P < 0.01). There is a positive relationship between kernel hardness and protein amount in some wheat cultivars [1]. As seen in Table 3, protein amounts of flour increased with the proportion of 8.30%, 8.86% and 11.80%, as the wheat hardness of samples increased. Thus, it is an estimated result that protein amounts of wheat samples increase, when their hardness increases.

Possibly, the applications decreasing flour ash amount in the tempering and milling processes also decreased the protein amount [17,93,99]. Because US application led to increasing some flour coming from central endosperm having less intensive but good quality proteins. It can be said that this situation will reflect on flour quality positively.

3.5. The effect of the soaking with ultrasound application in the tempering of wheat on some technological properties of flour

According to the results of ANOVA, the significant main variation sources subjected to Duncan's multiple range test (P < 0.05) and the results are summarized in Table 3. Wet gluten, gluten index and Zeleny sedimentation and falling number test results and their discussions are expressed as below.

3.5.1. Wet gluten amount

One of the most practical and easy methods to determine the quality of flour is wet gluten [82,100]. Gluten is responsible proteins for the dough formation of proteins. They affect rheological specifications of dough in the mixing and fermentation processes. It is very important for the bread industry in terms of estimation of rheological specifications such as flexibility, viscosity, extension ability [100,101]. As seen in Table 3, wet gluten amount increased, as kernel hardness increased. As the kernel hardness increases in the same wheat varieties, the amount of total protein and wet gluten generally increase [14,95,102,103]. Effects of tempering method on wet gluten were not statistically significant, possibly due to "Kernel Hardness $\,\times\,$ Tempering method" has a statistically significant effect on wet gluten amount (P < 0.01). But with a descriptive view, US application decreased gluten amount a little, as seen in the first study (Table 2). ANOVA results showed that the interaction of "Kernel Hardness × Tempering method" has significantly effect on wet gluten amount (P < 0.01). This interaction is given in Fig. 3, diagrammatically. This demonstration clearly shows the increase in wet gluten in wet gluten amount as kernel hardness of wheat sample increases, but being to be a little less for ultrasonic tempering than that of classic one especially for 45 and 65% hardness degrees. Here, it can be said that tempering with ultrasound application supplies flour having lower but high-quality protein from central endosperm [14], depending on water penetration depth into the endosperm by the effect of ultrasonication. Otherwise, it can be said what process causes physical changes in its structure during US application and loss of protein in wet gluten washing, especially for the lower hardness levels, not for 75% hardness level.

3.5.2. Gluten index

ANOVA results showed that only kernel hardness had significant effect on gluten index values at P < 0.05 level as being expected. But there was no significant effect of tempering method on the gluten index. As seen on the results of Duncan multiple comparison test (Table 3),

descriptively shown that, on the contrary to wet gluten amount, gluten index showed a little increase with US application, despite decrease in wet gluten amount. This phenomenon shows to be the increase in flour quality with the aid of US application in the tempering or the source of good quality proteins coming from the central endosperm (Table 3).

3.5.3. Zeleny sedimentation test

One of the very important parameters used to determine the wheat quality is Zeleny sedimentation test. It is considered as a criterion of protein amount and quality [82,104,105]. It was determined that the effects of kernel hardness and tempering method on Zeleny sedimentation value were significant. Duncan's multiple range test results showed that Zelenv sedimentation values increased considerably, while kernel hardness values of wheat samples increased. On the contrary, tempering with US applications decreased Zeleny sedimentation value like wet gluten. We need to glance at the physical and chemical specifications of wheat flours, obtained by tempering with ultrasound application. Possibly as a result of US application and better tempering, the flour coming from central endosperm increased and caused flour with lower but better quality protein, and also finer but lower starch damage at the flour below 140-µm granulation (Fig. 1). The interaction of "Kernel Hardness $\,\times\,$ Tempering Method", which was effective on Zeleny sedimentation values, was the same as on wet gluten as seen in Fig. 3. The same decrease was seen in the amount of ash and protein amounts, and wet gluten values. The decreasing of protein amount together with wet gluten and Zeleny sedimentation value were estimated results. Low starch damage and fine flour structure in granular structure, to which ultrasound application led, affected the decrease in Zeleny sedimentation value. This structural specification caused to increase in falling numbers. On the other side, the decrease in protein and ash amounts showed that the flour obtained by tempering with ultrasound applications comes from the central endosperm. These results may reflect wet gluten amount, as well as Zeleny sedimentation value [14,17].

3.5.4. Falling number

Falling number is an indicator of amylolytic activity. Falling number is considerably affected by the existence of kernel's amylase enzymes and damaged st amount of flour. In hard wheats, amylolytic activity is lower [82,93,105,106]. ANOVA results showed that the effect of kernel hardness proportion on the falling number was statistically important. According to Duncan's multiple comparison test results, falling number values increased as the kernel hardness of wheat samples increased (Table 3). Namely, amylolytic activity falls the contrary to an increase in kernel hardness [14,93,105–107].

Again, according to Duncan multiple comparison test results, tempering with ultrasound application led to an increase in falling number in comparison to classical one descriptively. We can explain this situation with the fact that ultrasound application increases the proportion of central endosperm parts in flour with the low enzyme activity and starch damage as a result of better tempering conditions.

3.6. The effect of ultrasound application in the tempering of wheat on rheological properties of dough

3.6.1. Farinograph experiments

3.6.1.1. Water absorption. According to ANOVA belonging to water absorption values of flour samples, it is determined that "Kernel Hardness" and "Tempering Method" variation sources have significant effect on water absorption. According to Duncan's multiple range test results (Table 4), water absorption values, especially in 75% hardness degree, increased together with the kernel hardness. This increment can be explained with the increase in wet gluten and protein amounts [14,95,102]. On the contrary, tempering with ultrasound application led to a decrease in water absorption values, in comparison to the classical one. There is a nearly 2.17% decrease.

		FARINOGRAM PROF	DERTIES			EXTENSOGRAM PR	DPERTIES		
Variation		Water absorption	Development time	Stability	Softening degree	Energy	Resistance to extensibility	Extensibility	Maximum Resistance
Sources	z	(%)	min.	min.	(BU)	(cm^2)	(BU)	(uuu)	(BU)
Kernel Hardness (%)									
45	4	$59.075^{b} \pm 1,50$	$1.675^{\rm b} \pm 0.23$	$7.200^{c} \pm 3,16$	$62.750^{a} \pm 11,55$	$50.750^{b} \pm 12,55$	$370.000^{b} \pm 129,16$	$93.250^{\rm b} \pm 1,70$	$370.500^{\mathrm{b}}\pm128,89$
65	4	$59.250^{b} \pm 0.54$	$1.750^{\rm b} \pm 0.33$	$8.450^{b} \pm 0.65$	$34.750^{b} \pm 4,19$	$68.000^{a} \pm 7,02$	$545.000^{a} \pm 70,13$	$101.500^{b} \pm 7,72$	$547.750^{a} \pm 71,96$
75	4	$62.250^{a} \pm 1.97$	$8.450^{a} \pm 0.71$	$17.100^{\mathrm{a}}\pm1,40$	$48.500^{ab} \pm 9.03$	$87.750^{a} \pm 9,74$	$442.250^{ab} \pm 31,85$	$126.000^{a} \pm 8,52$	$516.750^{a} \pm 40,63$
Tempering Method (%)									
Classic	9	$61.283^{a} \pm 2,08$	$3.883^{a} \pm 3,13$	$9.767^{\rm b} \pm 5,58$	$50.833^{a} \pm 17,78$	$62.167^{a} \pm 19,25$	$400.333^{\rm b} \pm 110,29$	$108.000^{a} \pm 12,99$	$420.500^{b} \pm 124,07$
Ultrasonic	9	$59.100^{b} \pm 1,32$	$4.033^{a} \pm 3,84$	$12.067^{a} \pm 4,43$	$46.500^{a} \pm 11,22$	$75.500^{a} \pm 15,88$	$504.500^{a} \pm 85,25$	$105.833^{a} \pm 19,33$	$536.167^{a} \pm 70,58$

Table

Ultrasonics - Sonochemistry 67 (2020) 105129

Possibly, due to low protein amount coming from central endosperm as a result of goo tempering in comparison to that of classic one (Table 3) as discussed before for the wet gluten and Zeleny values.

3.6.1.2. Development time. According to ANOVA results, it was determined that "Kernel Harness" at P < 0.01 level, also "Kernel Hardness \times Tempering Operation" interaction at P < 0.05 level have statistically affected dough development time. According to Duncan's multiple range test, dough development time increased as kernel hardness of wheat samples increase as being expected [14,82,88,108].

According to diagrammatic change, as seen in Fig. 4, the samples at 75% kernel hardness showed an excessive increase in dough development time, obtained from the flour obtained by tempering with US application, while it was affected contrarily a little in 45% and 65% hardness degrees. This increase for 75% hardness showed parallelism with protein, wet gluten and Zeleny values for tempering method (Fig. 3) due to the same causes discussed before. These results show that at the 75% hardness level, protein amount has a dominant effect on protein quality while at the lower hardness levels, protein quality has dominancy. It can be assumed that hard structural wheat gives more granular structure and more development time as a secondary factor [14,17,108,109,110].

3.6.1.3. Dough stability. According to ANOVA results, "Kernel Hardness" in P < 0.01 level, and "Tempering Operation" in P < 0.05 level have a statistical effect on stability. According to Duncan's multiple range test results, the dough stability of wheat samples increased together with their hardness degrees (Table 4). These values are respectively determined as 7.2 min, 8.45 min, and 17.1 min. Tempering with US application led to an important increase in dough stability value with 12,067% of farinogram in comparison to that of classical one with 9.767%. This data clearly shows that there is an increase in protein quality since obtained flour which has central endosperm sourced with high-quality protein by US application, even though there is a decrease in protein and wet gluten amount. The same progress was determined in the gluten index value. In protein distribution of sound wheat kernel, high-quality gluten which is important for bread making, takes place in central endosperm. The outer layer of floury endosperm has more intensive but less quality gluten than central zone. Gluten amount decreases towards central endosperm with quality increment [14,17,93,111].

3.6.1.4. Softening degree. According to ANOVA, "Kernel Hardness" at P < 0.01 level, interaction of "Kernel Hardness × Tempering Method" at P < 0.05 level, have a statistically significant effect on dough softening degree. As seen in Table 5, Duncan's multiple range test results clearly showed that as kernel hardness increased, a positive progress in dough softening degree in farinogram was observed, parallel to the increase in protein amount and quality as expected. With a descriptive view, a lower softening degree in farinogram ensued the tempering with US application (46.5 BU) in comparison to that of classical one (50.83 BU). It can be said that tempering with US application increases dough quality as explained in other qualitative developments, in comparison to those of classical one (Fig. 5). In Fig. 5, the interaction of "Kernel Hardness × Tempering Process" affects softening temperature (BU). As seen, on contrary to expectations, while dough softening degree would be the least at 75% hardness degree, it increased. The reason of this increase in softening degree for the 75% hardness level may be mechanical ripening of dough as a result of about three times long mixing time for longer dough developing and stability on farinograph than those of both lower hardness levels (Table 4). There is an opposite situation to the lower kernel hardness group in the 75%. This result can be explained with the fact that more granular structured flour obtained by tempering with US application gets more mechanic ripening of dough during kneading, as we already pointed out in our thesis in previous parts.

Table 5

The Changes in Some External and Internal Bread Properties of The Flour Obtained by Classic and Ultrasonic Tempering Methods as a Results of Duncan's Multiple Range Test ^{1.}

Variation Sources	N	Loafvolume (mL)	Spesificvolume (mL/g)	Crust color L*	a*	b*	Crumb color L^*	a*	b*
Kernel Hardness (%)									
45	4	$326.5^{a} \pm 19,07$	$2.367^{a} \pm 0,15$	$56.808^{a} \pm 2,28$	$14.328^{a} \pm 0,91$	$27.190^{a} \pm 0,79$	$65.373^{a} \pm 0,72$	$2.095^{ab} \pm 0.38$	$10.883^{a} \pm 5,45$
65	4	$333.0^{a} \pm 15,18$	$2.455^{a} \pm 0,16$	$58.385^{a} \pm 1,75$	$14.215^{a} \pm 1,15$	$24.983^{a} \pm 7,50$	$63.623^{a} \pm 2,71$	$2.007^{b} \pm 0,27$	$12.907^{a} \pm 0.89$
75	4	$354.0^{a} \pm 28,23$	$2.635^{a} \pm 0,17$	$58.802^{a} \pm 4,38$	$13.970^{a} \pm 1,59$	$32.430^{a} \pm 1,54$	$66.413^{a} \pm 1,65$	$2.782^{a} \pm 0,55$	$13.000^{a} \pm 1,18$
Tempering Method (%)									
Classic	6	$328.3^{a} \pm 19,49$	$2.397^{a} \pm 0,15$	$58.953^{a} \pm 3,22$	$14.003^{a} \pm 1,25$	$30.123^{a} \pm 3,15$	$64.532^{a} \pm 2,26$	$2.433^{a} \pm 0,65$	$13.633^{a} \pm 0,99$
Ultrasonic	6	347.3 ^a ± 23,85	$2.575^{a} \pm 0,19$	$57.043^{a} \pm 2,38$	$14.338^{a} \pm 1,15$	$26.278^{a} \pm 6,33$	$65.740^{a} \pm 1,88$	$2.157^{a} \pm 0.36$	$10.893^{a} \pm 3,99$

¹ The means with the same letter are not significantly different (P < 0.05)



Fig. 5. Interaction of "Kernel Hardness \times Tempering Method", which has effect on softening degree in pharynography.

3.6.2. Extensigraph experiments

3.6.2.1. Energy. According to ANOVA results, "Kernel Hardness" at P < 0.01 level and "Tempering Method" at P < 0.05 level have a statistical significant effect on the dough energy value. Duncan multiple range test results show that extensigram energy values increased, as kernel hardness of wheat samples increase. As expected increasing protein amount and quality together with hardness, enhanced energy value too. The flours that are obtained by the tempering method with US application from Bezostaya-1 samples, gave an increment in extensigram energy values in comparison to that of the classical one. In Table 4 positive effect of US tempering on dough energy values is shown clearly. When it is considered that extensigram energy values are a criterion to estimate protein amount and quality, the assumed facts for an increase in dough stability in farinograph and decrease in softening degree (Table 4) together with the increase in gluten index (Table 3) are valid for the change of energy value as well. In other words, we can say that US application in tempering process while causing a decrease in protein amount, increased the quality of protein coming from the central endosperm. As a result (Table 3 and 4), it can be concluded that bread quality will increase in concur with energy value increases [14,82,97,112] and accordingly, crumb texture gets better [14,17,112]. Hereby, it can be said that a mechanic ripening based on the granular structure of too hard wheat flours is more effective in the kneading process.

3.6.2.2. Resistance to extensibility. According to ANOVA results, "Kernel Hardness", "Tempering Method" and the interaction of "Kernel



Fig. 6. Interaction of "Kernel Hardness \times Tempering Operation", which has effect on resistance (BU) against elongation in extensigraph.

Hardness \times Tempering Method" have a statistically important effect on dough resistance to extensibility at P < 0.01 level. According to the interaction of "Kernel Hardness × Tempering Method", while the dough resistance obtained by classical tempering was 400.33 BU; the value obtained by tempering with US application increased to 504.50 BU. The interaction of the "Kernel Hardness \times Tempering Method", which had significant effect on dough resistance is given in Fig. 6. Here, there was a decrease in 75% hardness, while there was an increase in both wheat samples with 45 and 65% hardness. Tempering with US application increased the dough resistance of the samples with 45 and 65% hardness degree due to the flour coming from central endosperm with lower proteolytic activity and higher protein quality than those from the peripheral endosperm. The same situation didn't occur in 75% hardness degree, on the contrary, dough resistance decreased a little. Hereby, it can be said that a mechanic ripening based on the granular structure of too hard wheat flours is more effective in the kneading process.

3.6.2.3. Dough extensibility. ANOVA results belonging to dough extensibility data showed that wheat kernel hardness is statistically effective (P < 0.01). Duncan's multiple range test results showed that dough extensibility values increased together with resistance values, as the hardness values increased. These increases inboth dough resistance and extensibility positively reflected on dough energy values (Table 4). The effect of kernel hardness on dough extensibility is given in Table 5. Here, it can be said that the amounts of protein and wet gluten increasing with kernel hardness, positively affected the dough extensibility. But on contrary to this US application in tempering did

not significantly affect it (Table 4). As a result, the US application in tempering is more effective on dough resistance than those done on dough extensibility. The decreasing effect of US application in tempering on dough extensibility can be explained with the flour is sourced central endosperm, which has more protein strength and low proteolytic enzyme activity [14,113–115].

3.6.2.4. Maximum resistance. According to ANOVA results, the effect of "Kernel hardness" and "Tempering Method" are found statistically significant at P < 0.01 level. According to Duncan's multiple range test results, maximum resistance values show parallelism with the dough resistance to extensibility (Table 4). Tempering operation with ultrasound application increased maximum resistance value when compared to the classical one. Here, assumed reasons are valid for resistance against elongation as well. Also, the same discussions are valid for the interaction of "Kernel Hardness × Tempering Operation", found statistically significant in P < 0.01 level.

3.7. The effect of ultrasound application in the tempering process of wheat on bread making properties of flour

3.7.1. Loaf volume

According to ANOVA results, wheat kernel hardness and tempering method do not have a statistically important effect on bread volume due to a significant effect (P < 0.01) of their interaction. However, when Duncan multiple comparison test results are carefully examined, it is seen that bread volume increases with wheat kernel hardness and US application in tempering in comparison to those of classical one (Fig. 7). In Duncan test results, descriptively a qualitative superiority is perceived. As seen in "Kernel Hardness \times Tempering Method" interaction shown in Fig. 5, the increase in average bread volume values has superiority with increasing kernel hardness, and tempering with US application especially for 75%. It can be said that tempering with US application of the samples with 75% hardness degree in tempering are affected positively by the rise in gluten index (Table 4), development and stability times, softening degree at farinograph, and dough resistance to extensibility at extensigraph on loaf volume of the bread, despite the low protein and wet gluten values than those of classic method (Table 5). On the account of these results, the use of US applications in tempering causes to enhance flour and bread-making quality.



Fig. 7. Effect of "Kernel Hardness \times Tempering Method" intraction on loaf volume.

3.7.2. Specific volume

Specific volume of bread is used to estimate the staling tendency of bread. Puhr and D'Appolonia (1991), determined that bread's specific volume increases depending on water absorption ability [116,117]. Generally, the specific volume increases with protein amount and quality [14,95,118]. This is true for protein amount increasing with kernel hardness, but not for tempering method with US application. It is observed that the effect of "Kernel Hardness" and "Tempering operation" variation sources on specific volume values are not statistically important like those of loaf volume as a result of ANOVA. As seen Duncan multiple comparison test results (Table 5), with a descriptive assessment, specific volume values increased as kernel hardness increases, on the other side tempering with US application provided higher specific volume in comparison to the classical one. While the average specific volume value for bread, made from classical tempering applied wheat was 2.397, this value rose to 2.575 in bread made from the US applied samples. Discussions on specific volume changes showed parallelism with loaf volume.

3.7.3. Crust color

It was conducted, measuring L^* , a^* and b^* values of crust and crumb colors. Pinky-redness of crust color and whiteness of crumb are among important quality criteria. ANOVA results show that "Kernel Hardness" and "Tempering Method" variation sources do not have a statistically important effect on L^* , a^* ve b^* values. According to Duncan multiple range test results (Table 5), no meaningful result even descriptive was achieved in L^* and a^* values. When done a descriptive assessment, the b* values of bread's crust color increased with increasing kernel hardness. Kotancılar et al (2000) expressed that bread's crust color, made from weak wheat flour, was 10.7, and b* values of bread's crust color made from strong wheat flour, were 16.6, like our findings for kernel hardness [119]. This increment in yellowness of crust color may be coming from higher vellow pigment intensity or higher starch damage of hard kernel wheat as well known [120-122]. Also, the *b** value of the bread crust made from the wheat tempered with US application gave lighter crust color than that of the wheat tempered with the classic method. As a result, US application caused a more pinky crust color. Possibly, due to finer granulation and less amylolytic activity of flour obtained with US tempering method than those with the classic one.

3.7.4. Crumb color

According to ANOVA results, only "Kernel Hardness" among variation sources is found to be a statistically significant effect on a^* values of crumb color at P < 0.05 level. It is concluded that results arise from the intensive red color pigmentation of hard wheat than that of soft one [14]. When Duncan multiple comparison test result (Table 5) is descriptively examined, it is observed that all color parameters increased together with kernel hardness. Namely, the changes were observed in redness especially in crust color, and yellowness in crumb color values, despite no statistical importance. The increment in kernel hardness caused a more reddish crumb. The tempering with US application provided a whiter crumb color, possibly, due to enhancement in flour quality.

4. Conclusions

This research was conducted to show the effect of tempering with ultrasound (US) application in soaking of wheat having different kernel hardness, on kernel's water absorption, flour yield and flour and breadmaking qualities. The aim of the study at the industrial base, is to reduction two dampening stages of hard wheat to one in tempering, to decrease dampening time, and to enhance milling and bread-making quality. According to pre experiments test results, 30 s soaking period with US application showed the optimum time to increase water absorption of kernel 16% level for 75% kernel hardness degree. In industrial tempering applications, US application is considered to be more effective in tempering and may drop the dampening time below 20 s for semi-hard wheat kernel hardness. As a result, it is possible to decrease the dampening process from two stages to one. To industrial use, there are needs to develop industrial mechanization, application norms and flow diagram with new extra studies in the future. The flour yield and energy consumption were not affected by soaking with US application for 30 s possibly due to high experimental errors caused by low amount samples used for US application, but on the other side, it caused to increase in flour quality. Tempering with US application increased flour quality, by earning central endosperm sourced material, having flour from central endosperm with high quality but with less intensive gluten, due to deep penetration of water into central endosperm and high efficiency of tempering. These issues are approved by higher gluten index, lower ash amount, thinner flour granulation; and better dough rheology and ultimately higher bread-making test performance than those of classical tempering method. When it is considered from a scientific point of view, while tempering with US application speeded up water mass transfer into the wheat kernel, it caused to fine pixelization in central endosperm by cavitations in soaking with US application and the high speed and more effective tempering during rest time.

As a result, the use of the ultrasonic tempering method instead of classical one will drop the investment classical mechanization and operational expenses in milling technology. However, to get down the cost of the US apparatus and to design a new tempering system is the more priority of the flour milling industry.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.ultsonch.2020.105129.

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