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Effects of immature wheat on the nutritional and antinutritional quality of leavened and unleavened bread

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RESEARCH ARTICLE

Abstract

Three different wheat cultivars (Bezostaya-1, Gerek-79 and Kızıltan-98) were harvested at 2 maturity stages. Whole-wheat flour of immature wheat (WFIW) at 20% level and refined white flour of immature wheat (RFIW) at 10% level replaced commercial whole-wheat flour and refined white flour, respectively. Leavened and unleavened breads were prepared with those WFIW and RFIW flour blends. Some nutritional and antinutritional properties (ash, protein, fat, crude fibre, phytate phosphorus, phytic acid, minerals, total phenolic contents and antioxidant activity) of immature wheat and both types of bread were determined. The phytate phosphorus, phytic acid, total phenolic, P and K content of wheat decreased ($P<0.05$) with maturity. P, K, Fe and Zn content of bread prepared with Kızıltan-98 flour blends was found higher than that of other breads. The usage of WFIW at first maturity stage in leavened/unleavened bread preparation, increased the ash, total phenolic, mineral content and antioxidant activity compared to bread prepared with commercial whole-wheat flour. It was concluded that immature wheat especially at early stage of maturity is a rich source of nutrients for enriching leavened/unleavened bread prepared with WFIW flour blends.

Keywords: immature, phytic acid, minerals, total phenolic, antioxidant activity

1. Introduction

Wheat and wheat based foods are major source of nutrients which are mainly carbohydrates as well as protein, vitamins and minerals (Betschart, 1988). Besides, it contains phytochemicals that include tocopherols, tocotrienols, carotenoids, polyphenols, phytosterols and phytostanols which have additional health benefits (Tsao, 2008). Phenolic acids, flavonoids, tocopherols, tocotrienols and carotenoids are considered the major contributors of total antioxidant capacities of foods (Kim *et al.*, 2006; Yu *et al.*, 2004). These phytochemical antioxidants reduce the risks of coronary hearth deseases and cancer by neutralising excess free radicals, such as reactive oxygen species (Truswell, 2006; Tsao, 2008).

During the ripening period, not only physical propterties but also chemical composition of wheat kernel changes. The stage of maturity of wheat is monitored by different parameters, such as numbers of days after anthesis or moisture content of immature wheat. It was reported that moisture content at cutting is a better indicator of the degree of maturity than the number of days after anthesis (Tipples, 1980).

In the literature, the changes in the chemical composition of wheat were reported by different researchers. Mangels and Stoa (1928) harvested Marquis wheat at five different stages of maturity. They found that protein content of wheat showed no consistent variation at different stages of maturity for different seasons. They also reported that during maturation of wheat, the ash content and diastase activity of flour decreases. Skarsaune *et al.* (1970) reported that when the moisture content of two varieties of hard red spring wheat and two varieties of durum wheat decreased from 70 to 12% (30 days preripe to maturity), the total fat content reached a maximum value when the moisture content was near 65%, and then steadily declined. Nardi *et al.* (2003) analysed the quantities of simple sugars, fructans and aminoacids in immature grains of wheat, barley, rye and triticale. They reported that immature grains have a high content of lysine, fructans and simple sugars. The concentration of fructans and antioxidant compounds and vitamin C in wheat grains were significantly higher at early stage of kernel development and maturation, and then decreases rapidly 2-3 weeks after anthesis (De Gara *et al.,* 2003; Paradiso *et al.,* 2003). Due to the higher nutritional properties, immature wheat can be added to various cereal products to increase the nutritional status of foods.

In the literature, chemical properties of immature wheat were studied by numerous researchers but the effect of immature wheat on the nutritional/antinutritional properties of leavened and unleavened bread has not been studied in detail. Therefore, the aim of this study is to investigate the effect of immaturity on the nutritional/antinutritional quality of wheat, leavened and unleavened bread and to evaluate the effect of baking on nutritional/antinutritional quality of both types of bread.

2. Materials and methods

Wheat

Three different wheat cultivars (Bezostaya-1, Gerek-79 and Kızıltan-98) grown in Morcalı, Sarıkaya and Erenkavak-Anamas villages of Karaman, Turkey, were harvested at 2 different maturity stages according to the moisture content of the developing grains. First (earliest) harvest of Bezostoya-1, Gerek-79 and Kızıltan-98 are specified as BI, GI, KI and second harvest (more matured) as BII, GII and KII, respectively. The harvesting moisture content of BI, BII, GI, GII, KI and KII were 27.60, 18.89, 30.15, 21.32, 30.64 and 25.15%, respectively. Each wheat variety was harvested, tied as bunch and left in the field to air-dry at every stage of maturity. After passing through the threshing machines, samples were cleaned and stored at 4 °C until analysis.

Flour preparation

Wheat samples were milled on a hammer mill (Perten, LM3100, Perten Instruments AB, Huddinge, Sweden) to obtain whole-wheat flour with 100% extraction rate. To obtain refined white flour with 65±1% extraction rate, wheat samples were milled on a lab-roller flour mill (Chopin, CD 1, Villeneuve La Garenne, France).

Whole-wheat flour of immature wheat (WFIW) and refined white flour of immature wheat (RFIW) replaced commercial whole-wheat flour and commercial refined white flour. In our previous study, WFIW and RFIW replaced commercial whole-wheat flour (WFIW ratio; 0, 10, 20 and 30%) and commercial refined white flour (RFIW ratio; 0, 5, 10 and 15%) at four different ratios. Superior bread properties were obtained with 20% level of WFIW and 10% level of RFIW (Levent, 2014). Based on these results, a 20% replacement ratio for WFIW and 10% replacement ratio for RFIW was selected for the present study.

Bread preparation

For preparation of leavened control bread, 100 g flour, 3 g baker's yeast and 1.5 g salt are used as standard ingredients. Water is adjusted according to the Farinograph water absorbtion value. Homogeneous dough was prepared by a kneader (Hobart N50, Canada Inc., North York, ON, Canada). Fermentation times used for bulk fermentation were '30+30' min and for proofing 50 min. Fermented dough was baked in an oven (Arçelik ARMD-580, Istanbul, Turkey) at 250 °C for 12 min.

Unleavened control bread was prepared according to the method given by Başman and Köksel (2001): 200 g flour, 3 g salt and water based on Farinograph water absorbtion value were kneaded in a Hobart N50 mixer. After mixing, doughs were allowed to rest at 30 °C for half an hour and divided into four equal pieces, shaped like a ball, and sheeted by hand rolling to 1 mm thickness. After sheeting, they were baked on a preheated sac at 280±5 °C for 1 min.

Chemical analyses

The AACC methods were used for determination of moisture (method 44-19), ash (method 08-01), protein (method 46-12), fat (method 30-25) and crude fibre (method 32-10) (AACC, 1990). Phytic acid (PA) content of the samples was measured by a colorimetric method. PA in the sample was extracted using a solution of HCl (0.2 mol/l) and precipitated by an ammonium iron (III) sulphate dodecahydrate solution. For determining PA, phtate phosphorus value was multiplied by a factor 3.546 (Haug and Lantzsch, 1983).

For analysing the mineral contents, approximately 0.5 g of ground samples were put into a burning cup and mixed with 10 ml HNO_3 + H_2SO_4. The samples were incinerated in a microwave oven (MARS-5 Cem, Matthews, NC, USA) at 200 °C and dissolved ash was diluted to a certain volume with water. Contents were determined with an inductively coupled plasma atomic-emission spectrometer (Vista Series; Varian International AG, Zug, Switzerland) (Skujins, 1998).

Total phenolic content (TPC) were determined colorimetrically using Folin-Ciocalteu reagent as described by Paśko *et al.* (2009) with some modifications. The extracts were prepared according to the method of Chlopicka *et al.* (2012). Powdered samples (0.5 g) were extracted for 2 h with 10 ml solvent (methanol:HCl (0.16 mol/l):water, 8:1:1, v/v/v) at room temperature (25 °C). The extracts were separated by decantation and the residues were extracted again with 10 ml acetone (70 g/100 g) for 2 h and the extract was mixed with the initial methanol extract.

Table 1. Some chemical properties of wheat at two maturity stages.[1,2]

	Bezostaya-1		Gerek-79		Kızıltan-98	
Properties[3]	**Stage I**	**Stage II**	**Stage I**	**Stage II**	**Stage I**	**Stage II**
Ash (%)	1.65±0.03^{c}	1.64±0.01^{c}	1.58±0.05^{c}	1.46±0.02^{d}	1.92±0.04^{a}	1.78±0.01^{b}
Protein (%)	12.14±0.10^{b}	12.56±0.34^{b}	10.71±0.13^{c}	11.08±0.20^{c}	13.35±0.25^{a}	13.70±0.14^{a}
Fat (%)	1.98±0.11^{ab}	1.89±0.08^{ab}	2.04±0.06^{a}	1.94±0.03^{ab}	1.95±0.07^{ab}	1.82±0.03^{b}
Crude fibre (%)	2.62±0.17^{a}	2.30±0.14^{ab}	2.24±0.20^{ab}	2.10±0.14^{b}	2.35±0.16^{ab}	2.16±0.11^{b}
Phytate phosphorus (mg/100 g)	281.57±2.22^{c}	261.15±1.63^{e}	271.36±3.34^{d}	260.47±2.08^{e}	385.74±2.46^{a}	353.22±3.14^{b}
Phytic acid (mg/100 g)	998.44±4.86^{c}	926.05±8.55^{e}	962.24±5.98^{d}	923.61±7.93^{e}	1,367.84±4.02^{a}	1,252.53±5.70^{b}
TPC (µg GAE/g)	1,020.12±28.45^{a}	846.67±17.92^{b}	995.01±21.23^{a}	878.12±15.73^{b}	1,035.06±24.13^{a}	862.01±16.98^{b}
AA (% inhibition)	30.87±1.23^{a}	28.19±0.89^{bc}	28.90±1.26^{abc}	27.26±0.65^{c}	30.35±1.02^{ab}	29.65±0.92^{abc}
Mineral matter (mg/100 g)						
Ca	43.06±1.22^{ab}	42.22±0.31^{abc}	41.07±1.51^{bc}	39.45±0.64^{c}	44.74±1.04^{a}	41.78±1.95^{bc}
P	335.08±1.53^{c}	322.04±1.19^{e}	325.64±0.91^{d}	310.52±2.15^{f}	460.71±0.98^{a}	420.06±1.50^{b}
K	458.42±1.87^{b}	434.73±3.75^{d}	442.69±2.39^{c}	426.14±1.61^{e}	485.04±1.47^{a}	460.45±1.94^{b}
Cu	0.46±0.02^{b}	0.43±0.01^{bc}	0.42±0.03^{bc}	0.40±0.01^{c}	0.55±0.02^{a}	0.52±0.03^{a}
Fe	3.70±0.08^{c}	3.63±0.03^{cd}	3.62±0.06^{cd}	3.56±0.04^{d}	4.59±0.06^{a}	4.44±0.04^{b}
Zn	1.98±0.01^{b}	1.96±0.04^{b}	1.97±0.01^{b}	1.97±0.07^{b}	3.15±0.04^{a}	3.15±0.06^{a}

[1] Results are dry-weight basis.
[2] Means with different letters in the same row are significantly different ($P<0.05$).
[3] AA = antioxidant activity; GAE = gallic acid equivalent; TPC = total phenolic content.

For total phenolic assay, 0.3 ml aliquot extract was mixed with 2.7 ml de-ionised water, 0.3 ml Na_2CO_3 (20% w/w), and 0.15 ml Folin-Ciocalteu reagent. The absorbance was measured at 725 nm by using spectrometer (Hach, DR 5000, Düsseldorf, Germany). TPC was expressed as gallic acid equivalents (GAE).

Antioxidant activity (AA) was measured using a free radical 2,2 diphenyl-1-picrylhydrazyl (DPPH) solution in methanol according to the method given by Brand-Williams *et al.* (1995) with some modifications. Ground samples (1 g) were extracted with 10 ml methanol for 2 h and centrifuged at 3,000×*g* for 10 min. The supernatant (100 µl) was reacted with freshly made DPPH solution (3.9 ml, 25 mg/l) in methanol. The absorbance at 515 nm was measured at 0 and 30 min using a methanol blank. AA was calculated as % discolouration.

$$AA\% = [1-(\text{Abs sample}_{t=30}/\text{Abs control}_{t=0})] \times 100$$

Statistical analyses

The analyses of variance (ANOVA) was performed using the statistical software JMP 8.0 (SAS Institute, Cary, NC, USA). The comparison of the means was made using Student's t test. Significant differences were based on $P<0.05$.

3. Results and discussion

Some chemical properties of immature wheat are presented in Table 1. The ash content of Gerek-79 and Kızıltan-98 wheat decreased ($P<0.05$) with maturity. Compared to maturity stage I, fat, crude fibre and AA of wheat decreased at maturity stage II, but these changes were not statistically different ($P>0.05$). The results are in agreement with the earlier findings of Dexter and Matsuo (1977) and Skarsaune *et al.* (1970). The PA contents of immature wheat varied between 923.61 and 1,367.84 mg/100 g. Lolas *et al.* (1976) reported that the PA contents of 38 cultivars of wheat varied between 620 and 1,350 mg/100 g. Kızıltan-98 at first maturity stages has the highest, Bezostaya-1 and Gerek-79 at second maturity stages have the lowest phytate phosphorus and PA content among immature wheat cultivars. The PA content of wheat is effected by many factors, such as variety, climatic conditions, irrigation, soil, etc. (Bassiri and Nahapetian, 1977; García-Estepa *et al.*, 1999; Türksoy, 2005). Phytate phosphorus and PA value decreased with maturity in all wheat cultivars.

TPC and AA of immature wheat varied between 846.67 and 1,035.06 µg GAE/g, 27.26 and 30.87%, respectively. TPC of all wheat cultivars and AA of Bezostaya-1 decreased with maturation. Similarly, Xu *et al.* (2010) reported that maturation cause to decrease in TPC and AA of maize.

Ca, P, K, Cu, Fe and Zn contents of wheat samples varied between 39.45-44.74, 310.52-460.71, 426.14-485.04, 0.40-0.55, 3.56-4.59 and 1.96-3.15 mg/100 g, respectively. Demirbaş (2005) found that P, Cu, Fe and Zn contents of winter and spring wheat were 320 and 360 mg/100 g, 0.74 and 0.68 mg/100 g, 3.25 and 4.01 mg/100 g, 2.34 and 1.92 mg/100 g, respectively. P and K contents of all wheat cultivars, Ca and Fe contents of Kızıltan-98 decreased with maturity. Similarly, Inceer (2011) found that K, Mg, Mn, P and Zn contents of corn decreased with increasing maturity.

Some chemical properties of bread prepared from WFIW blends are presented in Table 2. The highest ash content was found in leavened bread prepared with Kızıltan-98 flour blend. In unleavened bread the ash content of bread prepared with Kızıltan-98 flour blend is significantly ($P<0.05$) higher than bread prepared from Gerek-79 flour blend. Elgün and Türker (2005) reported that the ash content of durum wheat flour is 25-50% higher than bread wheat flour in the same extraction value. The ash content of raw material may have influenced the ash content of both types of bread. Progress in maturity stage of wheat decreased the ash content of the leavened and unleavened bread. Also, bread prepared with maturity stage I flour blends had a higher ash content compared to control leavened and unleavened bread prepared with commercial whole-wheat flour.

The protein content of leavened and unleavened bread prepared from Kızıltan-98 blends was found significantly ($P<0.05$) higher than that of Gerek-79 flour blends. Durum wheat is known for its high protein content, which is 13.0% or higher (Halverson and Zeleny, 1988). Fat and crude fibre content of bread prepared with Bezostaya-1, Gerek-79 and Kızıltan-98 flour blends were not found significantly different from each other (Table 2). Also, the maturation stage of wheat did not change the protein, fat and crude fibre content of leavened and unleavened bread.

Phytate phosphorus and PA content of leavened and unleavened bread prepared from Kızıltan-98 flour blends were found significantly ($P<0.05$) higher than bread prepared from Bezostaya-1 and Gerek-79 flour blends. Average PA content of WFIW blends, leavened and unleavened bread were 902.42, 651.62 and 843.90 mg/100 g, respectively (data not shown for WFIW blends). Compared to WFIW blends, PA loss found in leavened and unleavened bread were 27.8 and 6.5%, respectively. It has been reported that PA content of wholemeal bread changed between

Table 2. Some chemical properties of bread prepared from whole-wheat flour of immature wheat blends.[1,2]

	n	Moisture (%)	Ash (%)[3]	Protein (%)[3]	Fat (%)[3]	CF (%)[3]	PP (mg/100 g)[3]	PA (mg/100 g)[3]	TPC (µg GAE/g)[3]	AA (% inhibition)[3]
Leavened bread										
Wheat cultivars										
Bezostaya-1	4	40.61a	2.82b	12.66ab	1.92a	2.11a	180.42b	639.92b	185.06a	10.73a
Gerek-79	4	41.00a	2.74c	12.40b	1.89a	2.06a	179.71b	637.69b	182.38a	10.41a
Kızıltan-98	4	40.27a	2.88a	12.91a	1.96a	2.08a	191.32a	677.26a	186.75a	10.60a
Maturity stage										
I	6	40.39a	2.85a	12.62a	1.93a	2.10a	185.10a	657.12a	185.87a	10.64a
II	6	40.86a	2.77b	12.70a	1.91a	2.06a	182.53a	646.14b	183.58a	10.52a
Control bread	2	41.10	2.82	12.75	1.94	2.04	181.20	642.54	180.04	10.44
Unleavened bread										
Wheat cultivars										
Bezostaya-1	4	8.97a	2.84ab	12.70ab	1.89a	2.08a	232.60b	826.29b	268.57a	11.41a
Gerek-79	4	9.13a	2.79b	12.35b	1.92a	2.03a	230.02b	817.46c	264.59a	11.21a
Kızıltan-98	4	9.08a	2.89a	12.99a	1.91a	2.06a	251.20a	887.95a	267.10a	11.48a
Maturity stage										
I	6	9.00a	2.87a	12.64a	1.93a	2.07a	240.05a	851.45a	268.13a	11.40a
II	6	9.12a	2.80b	12.71a	1.88a	2.03a	235.83a	836.35b	265.38a	11.34a
Control bread	2	8.80	2.85	12.68	1.91	2.08	229.57	814.07	260.07	11.27

[1] AA = antioxidant activity; CF = crude fibre; GAE = gallic acid equivalent; PA = phytic acid; PP = phytate phosphorus; TPC = total phenolic content.

[2] The means with the same letter in column are not significantly different ($P<0.05$).

[3] Results are dry-weight basis.

520 and 710 mg/100 g; 30-48% phytate loss was found in breadmaking. Also, phytate loss was determined in all stages of the bread making process (mixing, kneading, proving and baking). In the same study, 9% PA loss in unleavened wholemeal bread was found (McKenzie-Parnell and Davies, 1986). Effect of fermentation time and baking temperature on PA content of whole-wheat flour bread was studied by Qazi *et al.* (2003). While PA content of unleavened bread was 751.7 mg/100 g, PA content in leavened bread decreased to 661.1, 510.8 and 280.4 mg/100 g after 15, 30 and 45 min fermentation, respectively. Due to the phytase enzyme (naturally occurring in flour and yeast) and also the high baking temperature, phytate content of bread decreases in breadmaking (Plaami, 1997; Türk *et al.*, 1996). Similarly, progress in maturity stage of wheat decreased the PA content of the leavened and unleavened bread. Bread prepared with flour blends containing maturity stage I or II wheat had higher levels of PA compared to leavened and unleavened bread prepared with commercial whole-wheat flour.

TPC varied between 182.38 and 186.75 μg GAE/g for leavened and between 264.59 and 268.57 μg GAE/g for unleavened bread, respectively. TPC of leavened control bread (180.04 μg GAE/g) and unleavened control bread (260.07 μg GAE/g) were found lower than bread containing WFIW in formulation. TPC of leavened bread decreased by 78.6% when compared to TPC of WFIW blends (data not shown). Similarly, Leenhardt *et al.* (2006) reported that the phenolic content of bread decreased compared to flour and this may have been caused by interfering phenolic compounds during baking. The losses in TPC during dough mixing and kneading were reported by Chlopicka *et al.* (2012). However, Gélinas and McKinnon (2006) reported that baking caused an increase of TPC in bread. The formation of heat induced compounds from the Maillard reaction was reported as a possible contribution to the increased TPC. Compared with flour, the TPC loss in unleavened bread was 69%. This reduction was lower than that of leavened bread. This may have been caused by the shorter baking time of unleavened bread.

In the study conducted by Gujral *et al.* (2012), after germinating for 24 and 48 h, brown rice was milled into flour and incorporated in whole-wheat flour at a level of 10% for making chapatis. After baking the chapatti, the TPC decreased significantly in all blends. This decrease was 31% for the control bread and 3-29% in germinated and ungerminated brown rice added bread. The reduction in TPC after baking was attributed to heat effects on the flour components.

The AA of leavened and unleavened whole-wheat flour bread prepared from Bezostaya-1, Gerek-79 and Kızıltan-98 were (inhibition%) 10.73, 10.41, 10.60 and 11.41, 11.21 and 11.48 respectively. Bezostaya-1 and Kızıltan-98 flour blends increased the AA of leavened and unleavened bread compared to control bread. The average AA of WFIW blends, leavened and unleavened bread were 29.28, 10.58 and 11.37%, respectively (data not shown for WFIW blends). In leavened bread, AA loss was found as high as 63.8%. Losses of antioxidants during dough mixing, kneading and baking are reported by Leenhardt *et al.* (2006). Similarly, Chlopicka *et al.* (2012), reported that AA of control bread determined with FRAP and DPPH methods were decreased by 59.6 and 47.4%, respectively, according to wheat flour used in bread preparation.

P, K, Fe and Zn content of leavened and unleavened whole-wheat flour bread prepared with Kızıltan-98 flour blend were found to be superior than other bread (Table 3). Progress in maturity causes a decrease in P and K content of both leavened and unleavened whole-wheat flour bread (Table 3). Effects of the interaction between 'wheat cultivar × maturity stage' on the P content of leavened bread prepared from WFIW blends is presented in Figure 1. In all wheat cultivars, the P content of leavened bread prepared with WFIW blends was found higher at maturity stage I than at maturity stage II.

Ca, P, K, Cu, Fe and Zn content of leavened and unleavened bread prepared with commercial whole-wheat flour was increased by using WFIW blends, especially at maturity stage I. Similar results were observed in TPC and AA of leavened and unleavened bread (Table 3).

Some chemical properties of bread prepared with RFIW blends are given in Table 4. The moisture content of leavened bread prepared with RFIW blends was found between 38.03 and 38.18%. It has been reported that the moisture content is directly related with freshness of bread and ideal moisture content of bread should between 35 and 40% (Kamaliya and Kamaliya, 2001; Shah *et al.*, 2006). The ash content of leavened and unleavened bread prepared with Kızıltan-98 flour blend was found higher than that of other bread. The ash content of leavened and unleavened bread was decreased from 1.72 and 1.74 to 1.65 and 1.68%, respectively, with maturity. Bread (leavened and unleavened) prepared with flour blends containing maturity stage I had a higher level of ash compared to commercial refined wheat flour bread. Protein content of leavened bread was found between 11.44 and 11.78%. Also, protein content of leavened control bread prepared with commercial refined flour was found in this range. Protein content of unleavened bread prepared with Bezostaya-1, Gerek-79 and Kızıltan-98 flour blends was determined as 11.59, 11.36 and 11.69%, respectively. Similar to leavened and unleavened bread prepared with WFIW blends, the fat and crude fibre contents of bread prepared with RFIW blends were not found statistically different (P>0.05) from each other.

Table 3. Mineral contents of bread prepared from whole-wheat flour of immature wheat blends.[1,2]

	n	Ca	P	K	Cu	Fe	Zn
Leavened bread							
Wheat cultivars							
Bezostaya-1	4	53.04a	341.19b	481.25b	0.49a	3.95b	2.38b
Gerek-79	4	52.51a	339.40b	479.45b	0.48a	3.94b	2.36b
Kızıltan-98	4	53.38a	366.75a	486.21a	0.51a	4.14a	2.66a
Maturity stage							
I	6	53.29a	351.78a	484.40a	0.50a	4.02a	2.47a
II	6	52.66a	346.44b	480.21b	0.49a	4.00a	2.46a
Control bread	2	52.61	336.84	473.11	0.49	3.92	2.37
Unleavened bread							
Wheat cultivars							
Bezostaya-1	4	43.35a	363.73b	425.81b	0.44a	3.57b	2.05b
Gerek-79	4	42.84a	361.21c	422.48b	0.43a	3.54b	2.03b
Kızıltan-98	4	43.86a	389.95a	433.80a	0.46a	3.74a	2.27a
Maturity stage							
I	6	43.63a	374.25a	430.46a	0.45a	3.62a	2.12a
II	6	43.06a	369.01b	424.27b	0.43a	3.61a	2.11a
Control bread	2	43.09	358.18	418.66	0.44	3.54	2.06

[1] Means with different letters in the same column are significantly different (*P*<0.05).
[2] Results are dry-weight basis (mg/100 g).

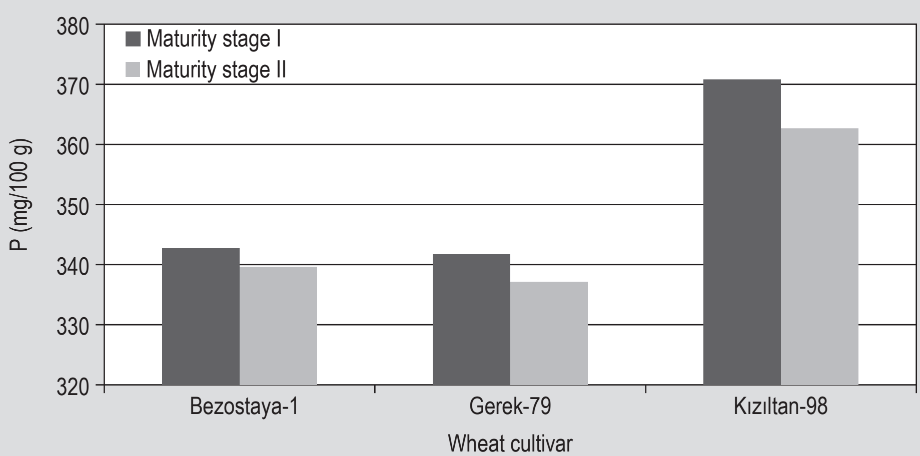

Figure 1. Effect of the interaction 'wheat cultivar × maturity stage' on the P content of leavened bread prepared from WFIW blends.

The average PA content of RFIW blends, leavened and unleavened bread was 185.65, 53.73 and 149.52 mg/100 g, respectively (data not shown for RFIW blends). Compared to RFIW blends, PA loss found in leavened and unleavened bread was 71.0 and 19.4%, respectively. McKenzie-Parnell and Davies (1986) reported that the phytate loss was 67-90% in white leavened bread. In another study, PA in various bread consumed in Iran were examined and it was found that Baggett bread, cylindrical or round bread used in fast food services, has the lowest and lavash bread has the highest PA content. It was claimed that due to sufficient fermentation processes or using low extracted flour Bagget bread has a low PA content (Gargari *et al.,* 2007).

TPC of leavened and unleavened bread prepared with RFIW blends was found to be lower than that whole-wheat flour bread. Gélinas and McKinnon (2006) reported that wholemeal bread was a much better source of phenolic compounds than white bread. Baking, due to the Maillard reaction products, slightly increased the concentration of

Table 4. Some chemical properties of bread prepared from refined white flour of immature wheat blends.[1,2]

	n	Moisture (%)	Ash (%)[3]	Protein (%)[3]	Fat (%)[3]	CF (%)[3]	PP (mg/100 g)[3]	PA (mg/100 g)[3]	TPC (µg GAE/g)[3]	AA (% inhibition)[3]
Leavened bread										
Wheat cultivars										
Bezostaya-1	4	38.06a	1.68b	11.69ab	0.78a	0.52a	15.17ab	52.90b	139.78a	5.68a
Gerek-79	4	38.04a	1.62b	11.44b	0.82a	0.52a	14.62b	51.89b	140.49a	5.60a
Kızıltan-98	4	38.18a	1.76a	11.78a	0.84a	0.50a	15.98a	56.42a	140.51a	5.66a
Maturity stage										
I	6	38.03a	1.72a	11.61a	0.84a	0.53a	15.60a	54.22a	141.60a	5.67a
II	6	38.16a	1.65b	11.66a	0.78a	0.50a	14.91a	53.25a	138.91a	5.62a
Control bread	2	37.45	1.70	11.56	0.80	0.51	14.78	52.42	140.10	5.62
Unleavened bread										
Wheat cultivars										
Bezostaya-1	4	8.12a	1.70b	11.59a	0.76a	0.50a	41.23b	146.12b	187.78a	6.04a
Gerek-79	4	7.93ab	1.65b	11.36a	0.82a	0.49a	40.72b	144.43b	189.11a	5.93a
Kızıltan-98	4	7.80b	1.78a	11.69a	0.70a	0.52a	44.62a	158.01a	188.60a	6.01a
Maturity stage										
I	6	7.92a	1.74a	11.53a	0.78a	0.51a	42.51a	150.53a	190.74a	6.01a
II	6	7.98a	1.68b	11.57a	0.80a	0.49a	41.87a	148.52a	186.25a	5.97a
Control bread	2	8.16	1.67	11.52	0.78	0.49	40.65	144.17	190.21	5.96

[1] AA = antioxidant activity; CF = crude fibre; GAE = gallic acid equivalent; PA = phytic acid; PP = phytate phosphorus; TPC = total phenolic content.
[2] The means with the same letter in column are not significantly different (P<0.05).
[3] Results are dry-weight basis.

phenolics in the bread crust and this was better seen in white bread than in wholemeal bread, which already contains high levels of phenolics. It was claimed that Maillard reaction products also have AA (Borreli *et al.,* 2003).

AA of leavened bread was found between 5.60 and 5.68%. AA of leavened control bread prepared with commercial refined flour was determined as 5.62%. The average AA of RFIW flour blends, leavened and unleavened bread was 8.68, 5.65 and 5.99%, respectively (data not shown for RFIW blends). AA of both types of bread decreased according to flour used in bread preparation. This decrease was 34.9% in leavened bread and 30.9% in unleavened bread prepared with RFIW blends. Peng *et al.* (2010) reported that AA of grape seed extract added bread decreased with thermal processing by around 30-40%. In contrast, Randhir *et al.* (2008) reported that thermal processing in general improved the TPC and AA in winter wheat.

Mineral content of bread prepared with RFIW blends is given in Table 5. The P, K and Zn content of leavened and unleavened bread prepared with Kızıltan-98 flour blend was found higher than bread prepared with Gerek-79 flour blends. P and K content of leavened and unleavened bread decreased (P<0.05) with maturity. According to the maturity stage variance source, the changes in moisture, protein, fat, crude fibre, phytate phosphorus, PA, TPC, AA, Ca, Cu, Fe and Zn content of leavened and unleavened bread were not statistically different (P>0.05) from each other (Table 4 and Table 5).

4. Conclusions

As a result of this study, it can be concluded that the use of immature wheat flour at first maturity stage increased the TPC, AA, Ca, P, K, Cu, Fe and Zn content of leavened and unleavened bread prepared with WFIW blends compared to commercial whole-wheat bread. TPC and AA of bread decreased by baking, but this decrease was less in unleavened bread comparing to leavened bread. However, PA content of unleavened bread was found higher than that of leavened bread. Further studies on other nutritional characteristics (fructooligosaccharides content, amino acid composition, etc.) of immature wheat harvested at earlier stages of maturity are needed to reveal the nutritional value of immature wheat.

Table 5. Mineral contents of bread prepared from refined white flour of immature wheat blends.[1,2]

	n	Ca	P	K	Cu	Fe	Zn
Leavened bread							
Wheat cultivars							
Bezostaya-1	4	28.13a	138.02a	193.18b	0.27a	1.40ab	1.14ab
Gerek-79	4	28.05a	126.76b	191.07b	0.27a	1.38b	1.09b
Kızıltan-98	4	28.23a	138.77a	203.65a	0.28a	1.44a	1.18a
Maturity stage							
I	6	28.19a	136.57a	198.38a	0.27a	1.41a	1.14a
II	6	28.06a	132.47b	193.55b	0.26a	1.40a	1.13a
Control bread	2	27.28	128.62	192.64	0.27	1.40	1.13
Unleavened bread							
Wheat cultivars							
Bezostaya-1	4	22.59a	137.52b	169.70b	0.26a	1.27b	0.88ab
Gerek-79	4	22.46a	136.18b	166.53c	0.24a	1.28a	0.84b
Kızıltan-98	4	22.74a	149.56a	177.58a	0.26a	1.33a	0.93a
Maturity stage							
I	6	22.68a	142.61a	173.08a	0.25a	1.30a	0.89a
II	6	22.51a	139.57b	169.46b	0.25a	1.29a	0.88a
Control bread	2	22.38	137.84	168.14	0.26	1.29	0.88

[1] Means with different letters in the same column are significantly different ($P<0.05$).
[2] Results are dry-weight basis (mg/100 g).

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