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Changes in the bioactivity of einkorn wheat during the maturation period and their effect on the properties of einkorn bread

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Abstract: Immature cereals may contain higher levels of nutrients than mature cereals. Immature and mature einkorn wheats were compared in terms of physicochemical, technological, rheological, and nutritional qualities and their performances for making bread were revealed. The values for thousand grain weight, hectoliter weight, sedimentation value and falling number of immature and mature einkorn wheat samples were 23.25–32.29 (g), 72.32–79.11 (kg/hL), 15.02–18.07 (mL) and 312–341 (sec), respectively ($p \leq 0.05$). Immature einkorn wholegrain flour (IEWF) contains more than twofold mineral substance than mature einkorn wholegrain flour (MEWF). The total phenolic content and antioxidant properties of IEWF were found to be higher than those of MEWF. The empirical rheological properties of IEWF were higher than those of MEWF except for degree of softening . The G′ of both IEWF and MEWF were always found higher than the G″, so they exhibited elastic behavior. The crust color brightness values of breads made from IEWF and MEWF were significantly different from bread made from commercial wheat wholegrain flour (CWF), but similar values were obtained for crumb color. The overall acceptability score of IEWF bread was similar to CWF bread, thus IEWF could be a good candidate for bread making or use in different food formulations.

Key words: Immature and mature einkorn wheat flours, rheological properties, bioactive properties, bread baking, sensorial evaluation

1. Introduction

Cereals have been one of the most important food sources for years, and among them the wheat varieties undoubtedly hold an important historic position. Demand for sustainable and healthy foods has increased since the 2000s, which has increased the attention paid to cereals like einkorn, spelt, and emmer (Keceli et al., 2021). Einkorn wheat (*Triticum monococcum* ssp. *monococcum* L.) , one of the ancient wheat varieties, has 14 chromosomes and is a close relative of durum (*Triticum turgidum* ssp. *durum*) and bread wheat (*Triticum aestivum* ssp. *aestivum*) (Hidalgo and Brandolini, 2014). Recent trends toward sustainable agriculture, along with an increasing focus on the nutritional value of food, have resulted in the rediscovery of various forgotten cereals, including einkorn (Hidalgo and Brandolini, 2014). Einkorn has exceptional nutritional qualities: a high protein content, high concentration of carotenoids, tocopherols, healthy lipids (mostly unsaturated fatty acids), high dietary fiber, trace minerals (iron and zinc), and low lipoxygenase activity (Hidalgo et al., 2016).

Many valuable compounds are lost with maturation, so immature cereals generally contain higher proportions of valuable compounds (Çetin-Babaoğlu et al., 2020). Nowadays, immature wheat has been attracted more attention because it contains less starch and more fiber than mature wheat (Çetin-Babaoğlu et al., 2020). Many studies have been carried out on the use of immature wheats in food formulations (Saa et al., 2017; Çetin-Babaoğlu et al., 2020). While adequate research has been done on the features of mature einkorn wheat, few in-depth studies have been conducted on the characteristics of immature einkorn and its use it in food formulations (Cankurtaran-Kömürcü and Bilgiçli, 2023; Kömürcü and Bilgiçli, 2023).

In this study, the physicochemical, nutritional and bioactive properties of immature einkorn (IE) and mature einkorn (ME) wheat grown in the same fields by farmers in the Kastamonu province of Türkiye were investigated, and bread production was carried out using both IE and ME wholegrain flours. An additional aim was to produce a bread from IE wholegrain flour with superior textural and rheological properties that would be acceptable in terms of sensory properties.

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2. Materials and methods

2.1. Harvesting einkorn wheat

Einkorn wheat was planted by local farmers in 4000 m^2 fields in Devrekani, Kastamonu, Türkiye. Since einkorn wheat has a dense stem structure and a single spikelet, it can be harvested from poor soils with low fertility and in dry and cold climates. Although climatic conditions can affect the physicochemical properties of soils, Baba (2021) documented that the soils of this particular region display a slightly alkaline pH (7.75), are unsalted, possess low organic matter content, and are moderately calcareous and loamy in nature. Additionally, the total nitrogen amount in the soil is moderate, while the phosphorus and potassium levels are high. When the einkorn grew into spikes, a part of the field was harvested as IE wheat at the milk-formation period. The remaining einkorn wheat was harvested as ME when fully mature. Both harvested einkorn wheats were dried in the field under natural sunlight. The IE and ME wheats used in this study were harvested from the same field but at different times.

2.2. Analysis of the immature and mature einkorn samples

2.2.1. Determination of the physical properties of the einkorn samples

The thousand grain weight (TGW) and hectoliter weight (HW) of the einkorn samples were determined according to Kulathunga et al. (2021). Briefly, the TGW of the IE and ME was determined by counting the grains in a 20 g, with the results were given as grams/dry matter.

2.2.2. Milling procedures of einkorn samples

The IE and ME wheats were dried under sunlight for seven days until reaching a moisture content of 11%–13 % and freed from husks and foreign materials. Wholegrain einkorn flours were obtained by grinding the IE and ME wheat in a blade mill (Millsan, Türkiye).

2.3. Physicochemical properties of the wholegrain einkorn flours

Moisture content, ash, and protein values of the wholegrain einkorn flours were determined according to standard methodologies as described by Ayvaz et al. (2021). Briefly, the moisture content of the flours was analyzed by weighing around 2 g of flour into tared crucibles, heating at 130 ± 1 °C for 1 h, and measuring the lost weight as the moisture content. To determine ash content, a known amount of flour was weighed into tared porcelain crucibles and burned in a muffle furnace at 550 °C for 6–8 h until white or gray ash was obtained. The ash content was determined by weighing the ash remaining at the end of combustion. The protein content of the flours was analyzed using the classical Kjeldahl protein determination method. The sedimentation and delayed sedimentation values, wet gluten (WG), gluten index (GI), and falling number (FN) of the wholegrain einkorn flours were measured according to Ayvaz et al. (2021). For the WG and GI values, the flours were weighed, placed in the right and left pouches of the gluten washing device, and washed with 2% salt water. The WG obtained from the gluten device was centrifuged, and the rotten and nonrotten parts were weighed. Finally, the WG (rotten gluten + nonrotten gluten) and GI (100 * rotten gluten / WG) values were calculated.

2.4. Mineral content of wholegrain einkorn flours

The Ca, K, Mg, Zn, and Fe contents of the wholegrain einkorn flours were determined by using an inductively coupled plasma-optical emission spectrometer (ICP-OES) (Turhan and Kurnaz, 2021). The preparation of the samples was carried out according to EPA method 3051A (microwave-assisted acid digestion of sediments, sludges, soils, and oils).

2.5. Resistant, nonresistant, and total starch and dietary fiber contents of wholegrain einkorn flours

The resistant, nonresistant, and total starch contents and the total dietary fiber contents of the IE and ME wholegrain flours were determined using a Megazyme resistant starch assay kit and a total dietary fiber assay kit; the analyses were performed according to the prescribed assay procedures (Megazyme, Ireland).

2.6. Morphological analysis of wholegrain einkorn flours

Scanning electron microscopy (SEM, Zeiss EVO® LS 10, Germany) was used to determine the morphological characteristics of the wholegrain einkorn flours. The wholegrain einkorn flours were examined under the microscope with magnifications of 1000× and 2500× using an accelerating voltage of 7 kV.

2.7. Empirical rheological measurement of wholegrain einkorn flours

The farinograph analyses of the samples were conducted a Brabender farinograph (Brabender, Germany) according to American Associates of Cereal Chemists International (AACCI) method 54-21 to determine the water absorption, development time, softening degree, and stability values of the IE and ME wholegrain flours (Kulathunga and Simsek, 2023).

2.8. Fundamental rheological measurement of wholegrain einkorn flours

The viscoelastic properties of the IE and ME wholegrain dough samples were defined using a rheometer (Anton Paar MCR 302, Austria) equipped with a peltier heating/cooling system (Upadhyay et al., 2012) and a PP25 parallel plate probe. An amplitude test was applied in the range of strain values between 0.01% and 100% to define the linear viscoelastic region. A stress sweep test was carried out at 0.1–10 Hz and 0.03% strain, and the gap spacing was set to 1 mm. After the probe reached the desired level, the samples sat for 3 min to allow residual stress relaxation.

With the frequency sweep test, storage modulus (G′) and loss modulus (G″) values were obtained as a function of angular frequency. The analysis was done in triplicate at 25 °C for each dough sample.

2.9. Determination of total phenolic content (TPC)

The TPC values of the wholegrain einkorn flour extracts were determined with a methodology specified by Singleton and Rossi (1965). Briefly, 30 mL of 80 % (v/v) methanol was added to 5 g of dried IE and ME wholegrain flours and shaken for 4 h at ambient temperature. The solution was filtered with a 0.45 µm filter and the extracts were preserved at –18 °C until the TPC analyses. For the analyses, 0.5 mL wholegrain einkorn flour extract, 2.5 mL folin reagent, and 2 mL Na_2CO_3 solution (7 % w/v) were mixed and left to sit for 30 min at ambient temperature. Absorbance values were determined using a spectrophotometer (Optizen Pop Bio Uv/Vis Spectrophotometer, Korea) at 765 nm. Results were given as mg GAE/kg DM. Analyses were carried out in triplicate.

2.10. Antioxidant capacity (AC) of wholegrain einkorn flours

The AC values of the einkorn wholegrain flours were determined using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) and copper-reducing antioxidant capacity (CUPRAC) methods. For the DPPH methodology, 0.1 mL wholegrain einkorn flour extract was added to 4.9 mL of DPPH solution (0.1 mM) and left to rest at ambient temperature for 20 min. Absorbance values were determined at 517 nm. Results were given as µmol TE/kg DM. Analyses were carried out in triplicate (Singh et al., 2002).

For the CUPRAC method, 0.1 mL of wholegrain einkorn flour extract was mixed with 1 mL each of $CuCI_2$, neocuproine, $C_2H_7NO_2$, and distilled water (Pekkirişci et al., 2023) and left to sit at ambient temperature for 60 min. Absorbance values were determined at 450 nm. Results were given as µmol TE/kg DM. Analyses were carried out in triplicate.

2.11. Determination of baking performance of wholegrain einkorn flours

2.11.1. Bread production method

The breads were made by following AACCI method 10-10.03 with small changes (Keceli et al., 2021). For the bread production, 60% (w/w) wholegrain einkorn flour, 2.5% (w/w) yeast (*Saccharomyces cerevisiae*), 1% (w/w) salt, and water 36.5% (w/w) were used. The mixture was kneaded for 5 min. The doughs were formed and fermented at 85% moisture and 30 °C; the initial and second aerations were performed at intervals of 30 min. Then, the doughs were placed into cooking bobbins and fermented for 30 min more in the same conditions before being baked for 25 min (Fimak, Türkiye) at a temperature of 235 °C. The bread production was carried out in three replicates.

2.11.2. Specific volume and texture analysis

The volumes of the bread samples were determined using the rapeseed displacement methodology (Ayvaz et al., 2021). The textural values of the bread samples were determined by using a texture device (TA.XD Plus, UK) provided with 5 kg load cell and a 36 mm diameter probe. The samples were compressed twice during the analysis. The breads were sliced into 2 × 2 × 2 cm cubes and the following conditions were used: test speed 1.7 mm/s, 30% compression, and wait time of 5 s between initial and second compression cycle. Finally, a time-dependent force graph was used to calculate the textural values.

2.11.3. Color parameters

The color parameters of brightness, redness, and yellowness (L^*, a^*, and b^*, respectively) of the bread samples were identified using a chromameter (CR-400, Konica, Japan) (Çakır et al., 2020). Briefly, these three values of the crumb and crust of each bread sample were determined from three distinct points. The results were then expressed as the average of three measurements per bread sample.

2.11.4. Sensory analysis

The sensorial assessment protocol was reviewed and approved by the Yıldız Technical University Food Engineering Department. The panelists were informed about the study and consent to participate in the study given by each panelist prior to the sensorial evaluation. The sensory characteristics of the breads made from immature and mature einkorn wholegrain flours were determined using the method suggested by Çakır et al. (2020) with some modifications. The sensory analysis was applied by 15 panelists who varied in age, gender, and income. The bread samples were cooled for 4 h after baking before being presented to the panelists for sensory analysis. The parameters, which were crust color, crumb color, pore structure, taste, odor, chewiness, and general taste, were scored between 0 and 10 (0 = extreme dislike, 10= extreme like).

2.12. Statistical analysis

The JMP 6.0 package (Statistical Discovery, LLC, SAS Institute) was used for the statistical analysis. All analyses were repeated thrice and presented as the average standard deviation of the replicates. To determine the significance of the discrepancies between the average values, a one-way analysis of variance was implemented.

3. Results and discussion

3.1. Physical properties of einkorn wheats

TGW is related to the milling value of wheat, and HW, which is used to determine the price of wheat, shows grain quality and grain bulk density (Kulathunga et al., 2021). TGW and HW values of the IE and ME wheats fell in the range of 23.25 ± 1.06 to 32.29 ± 1.58 g and 72.32 ± 3.08 to 79.11 ± 4.09 kg/hL, respectively. Only the difference between TGW values was found to be statistically important (Table 1, $p < 0.05$). High amounts of starch, protein and other nutrients accumulate during the maturation of the grains (Feng et al., 2021), so differences in TGW values may be related to maturation. Similar results were reported by Şaban and Ertop (2022), who found that the average TGW value of 20 different einkorn wheats was 30.93 g. Also, Kulathunga et al. (2021) reported that TGW values of einkorn wheat of 29.2 g.

3.2. Characteristics of einkorn wholegrain flours

IE and ME wholegrain wheat flours had the same moisture level (13 %), while the ash content of the IE (2.53%) was significantly higher than that of the ME (2.04%) (Table 1, $p < 0.05$). Similarly, Kim and Kim (2016) reported that the ash content of immature wheat kernels was higher than that of mature kernels. The protein content of the ME wholegrain flour (11.31 %) was significantly higher than that of the IE wholegrain flour (10.04 %) ($p < 0.05$). Kim and Kim (2016) also reported a higher protein content in mature wheat.

It is important to define the values of WG and GI of the einkorn flours to represent their bread producing capacity (Ayvaz et al., 2021). Fort he IE wholegrain flour, WG and GI values were not obtained due to its low viscoelastic properties. However, the WG and GI values of the ME wholegrain flour were 21.3 % and 52.0, respectively. The WG content of the ME was slightly below the 25% value required for bread (Piasecka-Jóźwiak et al., 2015). Ayvaz et al. (2021) reported the WG content of einkorn flour as 23%, and Piasecka-Jóźwiak et al. (2015) reported the WG values of four different einkorn flours as between 9.6% and 18%.

Another bread making quality indicator is the sedimentation value of cereal flours, and high gluten protein content is associated with a higher sedimentation value (Belcar et al., 2020). The sedimentation value of ME wholegrain flour was found to be 18.07 mL, which was statistically different from that of IE wholegrain flour, at 15.02 mL ($p < 0.05$). The desired sedimentation value for bread wheat should be at least 25 mL (Piasecka-Jóźwiak et al., 2015). Other studies reported varying sedimentation values for einkorn flour, with Izambaeva et al. (2016), Ayvaz et al. (2021) and Belcar et al. (2020) reporting 14.0 mL, 9.5 mL and 28.0 mL, respectively.

The FN shows α-amylase efficiency and is implicitly related to baking characteristics (Ayvaz et al., 2021). The FN values of the IE and ME wholegrain flours were determined to be 312 s and 341 s, respectively, which is a significant difference at $p < 0.05$. Varied FN value of einkorn flours were reported by Ayvaz et al. (2021), Belcar et al. (2020), and Izambaeva et al. (2016) as 340 s, 352 s and 374 s, respectively; Piasecka-Jóźwiak et al. (2015) also reported FN values of 4 different einkorn flours as 319–386 s. The ideal interval of FN is described as 200-350 s (Belcar et al., 2020), and for better α-amylase efficiency, it should not be high (Ayvaz et al., 2021). In this study, the FN values for both the ME and IE wholegrain flours were within the specified range, but the ME wholegrain flour was closely to the upper limit, so it may have low α-amylase efficiency like other einkorn flours mentioned above.

Finally, differences in protein content, WG, GI, sedimentation, and FN values can be explained by growth and agricultural applications and genotypic variation (Ayvaz et al., 2021), milling processes, applied methodologies and environmental conditions.

3.3. Mineral content of einkorn wholegrain flours

Einkorn is a cereal that contains high amounts of microelements (Hidalgo and Brandolini, 2014). The mineral content (K, Fe, Ca and Mg) of IE wholegrain flour (IEWF) was found to be significantly higher than that of ME wholegrain flour (MEWF) (Table 2, $p < 0.05$). The higher mineral content of IEWF may be related to its higher ash content, and the mineral content of wheat decreases with maturation (Pekkirişci et al., 2023). Compared to Turhan and Kurnaz (2021), this study found lower Zn, K, and Fe levels, but higher Ca and Mg levels. These differences may be due to wheat variety, climatic conditions and fertilization (Pekkirişci et al., 2023).

3.4. Resistant, nonresistant, and total starch contents of einkorn wholegrain flours

The formation, content, and composition of starch affect the quality of flour and bakery products (Hidalgo and

Table 1. Physicochemical properties of immature and mature einkorn wheats and wholegrain flours

	TGW (g)	HW (kg/hL)	Moisture (%)	Ash (%)	Protein (%)	Wet gluten (%)	Gluten index (%)	Sedimentation (mL)	Delayed sedimentation	Falling number (sec)
IE	23.25 ± 1.06^b	72.32 ± 3.08^a	13.0 ± 0.1^a	2.53 ± 0.08^a	10.04 ± 0.18^b	ND	ND	15.02^b	21.0 ± 1.41^a	312 ± 12^b
ME	32.29 ± 1.58^a	79.11 ± 4.09^a	13.0 ± 0.1^a	2.04 ± 0.02^b	11.31 ± 0.35^a	21.3 ± 0.28	52.0 ± 2.8	18.07^a	21.0 ± 1.41^a	341 ± 13^a

Different superscript letters show significant ($p < 0.05$) differences between samples. ND: Not detected; IE: Immature einkorn; ME: Mature einkorn.

Table 2. Mineral content, starch and dietary fiber content, empirical rheological properties, and bioactive properties of immature and mature einkorn wholegrain flours.

		IEWF	MEWF
Mineral content (mg/kg)	Zn	<5	<5
	K	4.152 ± 78^{a}	2.956 ± 63^{b}
	Fe	58.21 ± 2.65^{a}	31.6 ± 1.50^{b}
	Ca	1376.0 ± 46^{a}	361.1 ± 9.8^{b}
	Mg	1600.0 ± 51.0^{a}	774.7 ± 14.3^{b}
Starch and dietary Fiber content (g/100g)	Resistant starch	0.38 ± 0.01^{a}	0.13 ± 0.02^{b}
	Nonresistant starch	53.04 ± 0.87^{a}	48.93 ± 1.54^{b}
	Total starch	53.41 ± 0.87^{a}	49.05 ± 1.52^{b}
	Total dietary fiber	10.18 ± 0.30^{a}	8.31 ± 0.02^{b}
Empirical rheological properties	Water absorption (%)	60.9 ± 0.14^{a}	57.7 ± 0.28^{b}
	Development time (min.)	4.75 ± 0.07^{a}	2.6 ± 0.14^{b}
	Softening degree (FU)	35.5 ± 3.53^{b}	75.5 ± 2.12^{a}
	Stability (min.)	3.65 ± 0.07^{a}	1.80 ± 0.00^{b}
Bioactive properties	TPC (mg GAE/kg DM)	1165.24 ±13.24^{a}	947.86 ± 11.63^{b}
	DPPH (µmol TE/g DM)	10.11 ± 0.11^{a}	4.80 ± 0.33^{b}
	CUPRAC (µmol TE/g DM)	28.32 ± 1.30^{a}	22.38 ± 0.99^{b}

Different superscript letters show significant ($p < 0.05$) differences between samples. IEWF: Immature einkorn wholegrain flour; MEWF: Mature einkorn wholegrain flour.

Brandolini, 2014). The nonresistant starch content and total starch content of IEWF and MEWF were 53.04-48.93 g/100g and 53.41-49.05 g/100g, respectively, and these differences were statistically significant (Table 2, $p < 0.05$). Hidalgo and Brandolini (2014) reported that the average starch amount of 72 different einkorn flours was 50.5–71.4 g/100g, and these values were consistent with the total starch amount in our study. Immature grains have less starch than mature grains as free sugars are converted to starch during maturation (Çetin-Babaoğlu et al., 2020). However, this study found that the starch values of IEWF were significantly higher than those of MEWF.

Resistant starch is slow to be broken down during digestion, so foods with a high percentage of resistant starch exhibit lower glycemic index levels (Hidalgo and Brandolini, 2014). Einkorn flour has a lower resistant starch level than bread wheat flour, at 2.56 g/100g and 3–8.8 g/100g, respectively (Hidalgo and Brandolini, 2014). The resistant starch levels of IEWF and MEWF in this study were 0.38 g/100g and 0.13 g/100g, respectively, and were lower than the values stated in the literature for both IEWF and MEWF.

3.5. Dietary fiber content of einkorn wholegrain flours

The dietary fiber contents of IEWF and MEWF were found to be 10.18 ± 0.30 g/100g and 8.31 ± 0.02 g/100 g, respectively (Table 2, $p < 0.05$). Kulathunga and Simsek (2022) reported the insoluble dietary fiber content of wholegrain einkorn flour as 7.53%. Hidalgo and Brandolini (2014) stated that the dietary fiber content of einkorn wheat is lower compared to bread wheat and durum wheat. Genotype, growth factors, geographic location, and method of analysis can all cause variation in dietary fiber content (Kulathunga and Simsek, 2022). Although IEWF stands out with its high fiber content in this study, the high fiber content of both IEWF and MEWF showed that they could be potential prebiotic nominees for new food products.

3.6. Microstructural properties of einkorn wholegrain flours

It has been observed that starch granules (large oval shapes) and protein bodies (small circles around starch) begin to form in IEWF, and the protein structures form a network in the endosperm (Figure 1a and 1b). On the other hand, in MEWF, starch granules and protein bodies were seen to mature so that the protein was embedded into starch granules (Figure 1c and 1d). Kulathunga et al. (2021) reported that there are two types of starch granules in bread wheat endosperm, the large (10–35 µm) A type and the small (1–10 µm) B-type. These two different types of starch granules were also detected in the MEWF in this study (Figure 1).

3.7. Empirical rheological properties of einkorn wholegrain flours

All farinograph parameters investigated in this study showed significant differences between IEWF and MEWF (Table 2). The water absorption of IEWF (60.9%) was higher than that of MEWF (57.7%) ($p < 0.05$). For optimum consistency, the desired water ratio for bread dough is around 65%; when it is below this value, the mixing time is prolonged (Biel et al., 2021). There is a close relationship between water absorption and amount of protein (Biel et al., 2021) and/or starch content (Kulathunga and Simsek, 2023; Pekkirişci et al., 2023); as the amount of protein increases, the amount of water absorbed by the flour increases (Biel et al., 2021). The higher water absorption values of IEWF than MEWF may be associated with higher total starch content and/or protein content.

The development time provides information on the mixing needs required for dough to reach maximum consistency (Kulathunga and Simsek, 2023). In this study, IEWF had an approximately two-fold longer development time than MEWF, at 4.75 min and 2.60 min, respectively. Varying results have been reported for einkorn flour dough development times. Wieser et al. (2009), Kulathunga and Simsek (2023) and Biel et al. (2021) reported dough development times of einkorn flour as 1.8–4.5, 1.8–2.0 and 2.1 minutes, respectively.

The degree of softening is defined as the loss of viscosity of the dough colloidal systems at certain times of kneading (Keceli et al., 2021). Dough with high softening level are less resistant to long mechanical processes (Biel et al., 2021). While this study found the softening degree of IEWF (35.5, FU) was lower than MEWF (75.5, FU), Biel et al. (2021) and Keceli et al. (2021) reported higher softening values for einkorn samples.

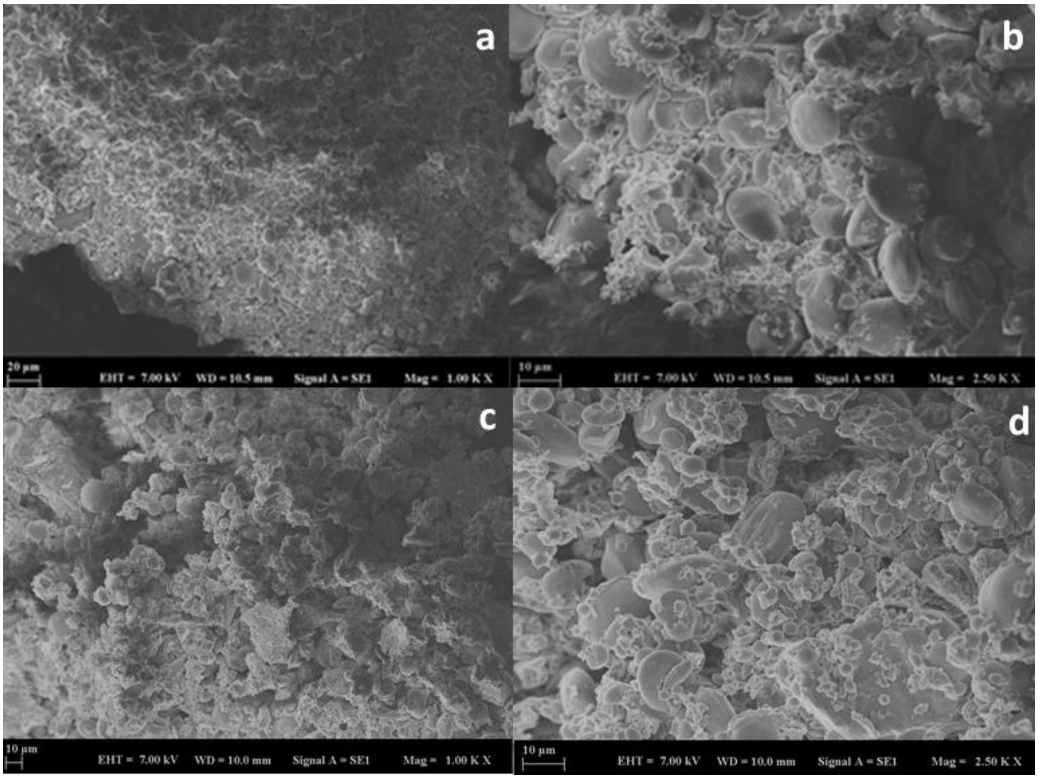


Figure 1. SEM (Scanning Electron Microscope) images of einkorn flours a; Immature einkorn whole grain flour (IE W F) 1000 x b; Immature einkorn whole grain flour (IE W F) 2500 x c; Mature einkorn whole grain flour (ME W F) 1000 x d; Mature einkorn whole grain flour (ME W F) 2500 x

A strong dough needs high stability values (Biel et al., 2021). In the present study, IEWF (3.65 min) showed higher a stability value than MEWF (1.80 min). This shows that IEWF dough has a more resistant structure against kneading. Kulathunga and Simsek (2023) reported the stability values of three different einkorn flours as 0.8, 0.7, and 0.7 min, which are significantly lower than those of emmer and spelt flours. In another study, Biel et al. (2021) found the stability values of einkorn, emmer, spelt, and common wheat flours to be 1.2 min, 1.43 min, 4.15 min and 4.50 min., respectively.

3.8. Fundamental rheological properties

The viscous and elastic behavior of the immature and mature einkorn wholegrain flours were determined with frequency sweep tests in the linear viscoelastic region (0.03% strain). G′ and G″ describe the dynamic features of dough; if G′ has a higher value than G″, the dough exhibits elastic behavior (Şerban et al., 2023). Since the G′ values of both IEWF and MEWF in this study were always higher than G″ values, both IEWF and MEWF showed elastic behavior (Figure 2). This was caused by the damping factor (tan δ (G″/ G′)) being less than one. Maturation significantly affected the G′ and G″ values of einkorn flours, particularly G′. Brandolini et al. (2023) and Şerban et al. (2023) stated that einkorn dough exhibited the same elastic behavior as found in this study, and the maturation affected the dynamic rheological characteristics of the einkorn flour doughs.

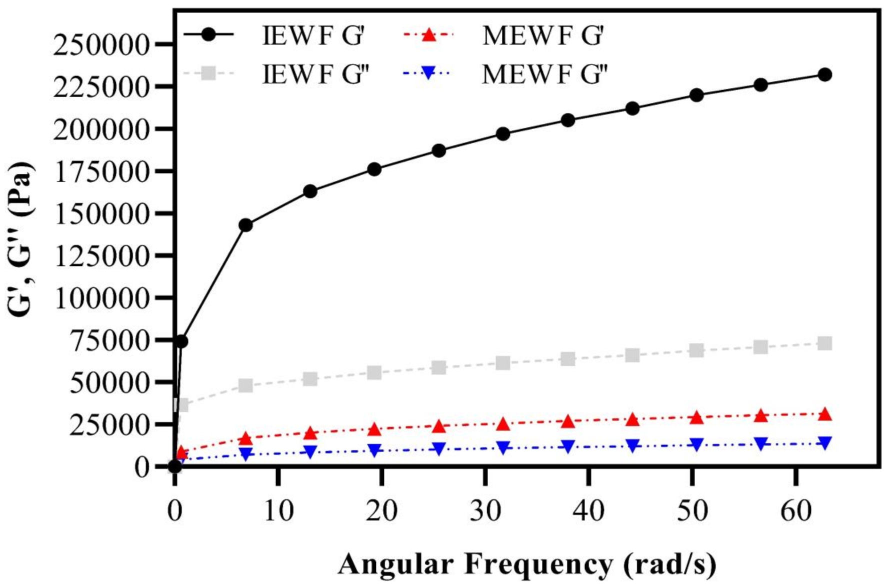
3.9. TPC and AC of einkorn wholegrain flours

The TPC values of IEWF and MEWF are presented in Table 2. The TPC values of IEWF (1165.24, mg GAE/kg DM) were found to be significantly higher than those of MEWF (947.86, mg GAE/kg DM) ($p < 0.05$). Kim and Kim (2016) reported that the TPC value of immature wheat grains is higher than that of mature wheat grains. However, Saa et al. (2017) reported that the TPC value of mature wheat flour is higher than that of unripe wheat. These differences in the literature may be related to wheat variety and/or the presence of phenolic compounds in bound or free form (Kim and Kim, 2016).

The DPPH/CUPRAC antioxidant activity results of IEWF and MEWF were 10.11/4.80 µmol TE/g DM and 28.32/22.38 µmol TE/g DM, respectively (Table 2). While the CUPRAC results were higher than the DPPH results, both revealed that IEWF had significantly higher antioxidant properties than MEWF ($p < 0.05$). Izambaeva et al. (2016) reported that the CUPRAC antioxidant activity of whole meal einkorn flour was 201.12 mMTE/100g, but they could not determine the DPPH antioxidant activity. Pekkirişci et al. (2023) reported the DPPH and CUPRAC values for einkorn bulgur as 335.68 mg TE/kg and 4.48 µmol TE/g, respectively. Kim and Kim (2016) reported that immature wheat grains have higher antioxidant activity than mature wheat grains.

3.10. Specific volume and textural properties of bread samples

Consumers typically select bread with a high specific volume (Ayvaz et al., 2021). The measured specific volume



IEWF: Immature einkorn wholegrain flour, MEWF: Mature einkorn wholegrain flour

Figure 2. Viscoelastic properties of the einkorn flours

values of commercial wholegrain flour bread (CWB), immature einkorn wholegrain flour bread (IEB), and mature einkorn wholegrain flour bread (MEB) were 1.73 mL/g, 1.72 mL/g, and 1.68 mL/g, respectively (Table 3, $p > 0.05$). Brandolini et al. (2023) reported the specific volume values for two different einkorn breads as 5.23 mL/g and 5.40 mL/g, both higher than that of their control bread. Çakır et al. (2020) found the specific volume of einkorn sourdough bread as 1.37 mL/g and stated that the specific volume of bread enriched with einkorn flour decreased.

The hardness and elasticity values of the IEB and MEB samples were found as 6.59 and 6.61 N and 0.42 and 0.40, respectively, both higher than the hardness and elasticity values of CWB (4.78N and 0.36) (Table 3, $p > 0.05$). Chewiness results of the IEB, MEB and CWB were 4.65, 4.26 and 3.06 N, respectively, and the only statistical difference was between the IEB and CWB samples ($p < 0.05$). Brandolini et al. (2023) stated that the hardness and elasticity values of bread produced from bread wheat flour were significantly higher than bread produced from two different einkorn flours. However, Çakır et al. (2020) emphasized that the hardness value of bread produced from einkorn flour was 15.69 N and that enrichment with einkorn flour in bread production increased the hardness value of the bread. Kömürcü and Bilgiçli (2023) stated that the use of ancient wheats such as einkorn and emmer in the production of bread increased the hardness and chewiness of the bread. Differences in the textural values of the samples can be attributed to the starch content of flours (Brandolini et al., 2023).

3.11. Color characteristics

The crust color L* value of CWB was higher than that of IEB and MEB, at 62.67, 51.85 and 57.57, respectively (Table 3, $p < 0.05$). The use of immature einkorn flour in bread production caused an *a** decrease in the brightness value of the crust color of the bread, so a darker color formation. The *a** values of the crusts of the produced breads were 2.47, 8.46, and 3.66 for CWB, IEB and MEB, respectively ($p < 0.05$). However, the use of immature einkorn wholegrain flour in bread production caused an increase in the redness value of the crust color of the bread. The crumb color values (*L**, *a** and *b**) of three different bread types are similar to each other and no statistical difference was found, except for in the b value of CWB ($p < 0.05$). Brandolini et al. (2023) found the crust and crumb *L** values of bread produced with two different einkorn flours and bread wheat to be similar. Çakır et al. (2020) reported that the crust and crumb brightness values of breads enriched with einkorn flour resulted in darker brightness values. The darker crust colors and increased redness values of IEB and MEB in this study are probably related to non-enzymatic browning reactions that occur during baking (Kömürcü and Bilgiçli, 2023). In addition, the fact that the *b** values of bread produced from einkorn flour were higher than those produced from commercial flour is related to the lutein pigment content, which is responsible for the yellow color of wheat (Kömürcü and Bilgiçli, 2023).

3.12. Sensorial properties

The sensorial properties are presented in Table 4. The breads with the highest scores for crust and crumb color were IEB, CWB, and MEB, respectively. The crust color values of IEB and CWB were not statistically significant, but these two breads were statistically different from MEB. In terms of crumb color values, IEB was found to be significantly different from CWB and MEB. Color is an important parameter in consumer acceptance, so IEB's color values being similar to or higher than those of CWB reveal its importance in consumer acceptance. The pore structure of IEB and CWB were similar and they both had better pore structure scores than MEB.

The taste, odor, and chewiness scores of CWB were statistically higher than IEB and MEB ($p \leq 0.05$). Although the taste and smell scores of the IEB were statistically

Table 3. Specific volume, texture profiles and color characteristics of einkorn bread samples.

		CWB	IEB	MEB
	Specific volume (mL/g)	1.73 ± 0.5^{a}	1.72 ± 1.3^{a}	1.68 ± 1.3^{a}
Textural profiles	**Hardness (N)**	4.78 ± 0.30^{a}	6.59 ± 0.77^{a}	6.61 ± 0.68^{a}
	Chewiness	3.06 ± 0.34^{b}	4.65 ± 0.45^{a}	4.26 ± 0.05^{a,b}
	Elasticity	0.36 ± 0.01^{a}	0.42 ± 0.03^{a}	0.40 ± 0.02^{a}
Bread crust color	**L***	62.67 ± 0.99^{a}	51.85 ± 0.97^{c}	57.57 ± 1.08^{b}
	a*	2.47 ± 0.13^{c}	8.46 ± 0.60^{a}	3.66 ± 0.36^{b}
	b*	25.18 ± 3.21^{a}	26.08 ± 0.62^{a}	28.68 ± 0.11^{a}
Bread crumb color	**L***	57.39 ± 2.46^{a}	55.56 ± 0.81^{a}	55.58 ± 0.18^{a}
	a*	2.53 ± 0.13^{a}	2.21 ± 0.61^{a}	2.19 ± 0.27^{a}
	b*	19.78 ± 0.04^{b}	25.23 ± 0.68^{a}	25.75 ± 0.24^{a}

Different superscript letters show significant ($p < 0.05$) differences between samples. CWB: Commercial wheat wholegrain flour bread; IEB: Immature einkorn wholegrain flour bread; MEB: Mature einkorn wholegrain flour bread.

Table 4. Sensory properties of einkorn bread samples.

Bread type	Crust color	Crumb color	Pore structure	Taste	Odor	Chewiness	General appreciation
CWB	5.10 ± 0.74[a]	5.00 ± 0.82[b]	6.40 ± 0.52[a]	5.40 ± 0.52[a]	5.40 ± 0.52[a]	5.60 ± 0.52[a]	5.30 ± 0.48[a]
IEB	5.80 ± 0.63[a]	6.30 ± 0.67[a]	6.90 ± 0.74[a]	3.10 ± 1.20[b]	4.00 ± 0.82[b]	3.40 ± 1.51[b]	4.60 ± 0.84[a]
MEB	4.40 ± 0.52[b]	4.20 ± 1.03[b]	5.40 ± 0.52[b]	2.50 ± 0.85[b]	2.60 ± 0.84[c]	2.40 ± 0.84[b]	3.60 ± 0.84[b]

Different superscript letters show significant ($p < 0.05$) differences between samples. CWB: Commercial wheat wholegrain flour bread; IEB: Immature einkorn wholegrain flour bread; MEB: Mature einkorn wholegrain flour bread.

similar to the MEB, the IEB had higher scores than the MEB in terms of taste, smell, and chewiness. CWB was the most popular type of bread in terms of important sensory parameters such as taste, smell, and chewiness, and IEB was the second most preferred bread type.

The highest and lowest general appreciation scores were applied to CWB and MEB, respectively. Also, the general appreciation scores of CWB and IEB were found to be significantly different than those of MEB ($p \leq 0.05$). These results showed that IEB has a similar general evaluation score with the CWB at a level that can be accepted sensorially. Çakır et al. (2020) reported that breads enriched with 25 % and 50 % einkorn flour had similar sensory results to thier control. In this study, the sensory scores of breads produced from IE flour were close to the control, which is promising for further studies on the use of einkorn flour in bread production.

4. Conclusion

The study findings indicate that IEWF has a higher mineral content and more dietary fiber than MEWF. Additionally, the bioactive and empirical rheological properties of IEWF are superior to those of MEWF. IEWF exhibited superior characteristics for bread manufacturing compared to MEWF, particularly with regards to rheological, textural, and sensory characteristics. Finally, this study demonstrates that using IEWF in the production of bread and other food products has the potential to improve both their nutritional value and sensory characteristics.

Conflict of interest

The authors report no conflict of interest.

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Availability of data

The study data can be provided by the corresponding author upon acceptable demand.

Author contributions

Mehmet Fatih Erkölencik: Methodology; Burcu Kahraman: Investigation; Görkem Özülkü: Investigation; Eray Tulukçu: Methodology; Hamza Göktaş: Writing, original draft; Osman Sağdıç: Methodology and investigation; Muhammet Arıcı: Project administration and supervision.

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