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A comparison study of ancient and modern wheat: Characterization and sensory analysis of an ancient Anatolian wheat Triticum dicoccum (Kavilca) grown in Kars, Türkiye

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Abstract

The ancient wheat varieties have been ignored due to efforts to increase the yield of modern wheat. In recent years, interest in ancient wheat has increased for various reasons. This study aimed to characterize the ancient Anatolian wheat *Triticum turgidum* ssp. *dicoccum* (Kavilca) from Kars, Türkiye and to compare it with modern bread wheat *Triticum aestivum* in terms of quality parameters, mineral and fatty acid profile, total flavonoid content (TFC), and antioxidant activity (TAA). Quality parameters were conducted with standardized methods. Mineral and fatty acid profiles were determined with inductively coupled plasma mass spectrometry and gas chromatography, respectively. Spectrophotometric methods were used to determine TFC and TAA. Triangular test and Quantitative Descriptive Profile method were applied for sensory analyses. Novel findings of this study were low gluten quality ($p < 0.05$), low Al content (2.85–5.16 mg/kg dw, $p < 0.05$), and high TFC (3.65 mg RE/g dw, $p < 0.05$) of Kavilca along with similar overall liking sensory score ($p > 0.05$) compared to modern bread. There was a strong sensory difference with a 99.9% probability between modern and 15% Kavilca added bread. Characterization and especially, sensory analysis results might be a reference for the food industry to integrate Kavilca into several bakery products.

1. Introduction

Wheat is a strategically important plant with a history of more than 10000 years and provides approximately 20% of the energy need of the world population. Türkiye, which is one of the important centers in terms of plant gene resources, is also the homeland of wheat, with its southeast included in the "Fertile Crescent". However, in the last 50 years, efforts to increase the yield and economic success of wheat have caused local and ancient wheat varieties to be forgotten and on the verge of extinction.

The cultivation area of ancient wheat varieties is gradually decreasing in Türkiye. However, in recent years, thanks to developing production systems and new trends in consumer demand, ancient wheat has come to the fore in the food industry. Local seeds reduce the foreign dependency of a country in terms of the agricultural economy by saving the farmers from the obligation to depend on international companies. Furthermore, plant genetic resources can be an important solution to feed the world population, reaching 10 billion in 2050 [1]. In addition, the increase in interest in organic products and the search for new food products have increased interest in local seeds. Today, as a result of a sedentary lifestyle and incorrect eating habits, cardiovascular, digestive system, and intestinal diseases, obesity and diabetes are common in societies. Ancient wheat attracts the attention of both producers and consumers due to its capacity to prevent chronic diseases due to its high protein, high antioxidant, and high fiber content. Ancient wheat varieties do not need pesticides and fertilizers, and they cannot be grown without organic and good farming practices; therefore, food safety is ensured. Sustainable agricultural practices, in addition to maintaining productivity in agriculture, reduce damage to the environment and increase the quality of life of farmers.

An Emmer-type wheat *Triticum turgidum* ssp. *dicoccum* (2n = 28 chromosomes), is a common crop in Kars, Türkiye, and is named “kavilca” and “kabluca” locally. It is known as rich in protein (17–19%) and fiber but low in gluten. The integration of *T. dicoccum* into the modern food industry is crucial; however, the characterization and information about the techno-functional and quality properties of Kavilca are needed to be enlightened by comparing the modern wheat because quality parameters and techno-functional properties are of great importance for processing and applications as an ingredient in foods. Previously, Akman and Karaduman [2] studied the quality characteristics of some *Triticum* species grown in rained and irrigated conditions. In this study, Kavilca was grown by local farmers in Kars known for its harsh climate conditions which is the homeland of Kavilca. Furthermore, to date, it has not been proven yet whether ancient wheat was healthier than modern wheat or not, and more types of ancient wheat needed to be studied [3]. No study to date has investigated the sensory profile of Kavilca bread from Kars, Türkiye. In this study, we aimed to characterize the ancient Anatolian wheat *T. turgidum* ssp. *dicoccum* (Kavilca) from Kars, Türkiye, and to compare it with the modern bread wheat *T. aestivum* in terms of quality parameters, techno-functional properties, mineral profile, fatty acid profile, total flavonoid, and antioxidant activity. In addition, to provide information about consumer preferences, we conducted a triangle test between 15% Kavilca added bread and modern bread along with a sensory descriptive test of 100% Kavilca bread, 15% Kavilca bread, and modern bread.

2. Materials And Method

2.1. Materials

T. dicoccum (Kavilca) was harvested and mixed from different fields of Arpaçay district of Kars Province, Türkiye. The ground whole grain flour of *T. dicoccum* and Esperia variety of *T. aestivum* were supplied and delivered to us by UNMAŞ Bakery Products. A fatty acid methyl ester (FAME) standard mixture (37 components) was purchased from Supelco (Bellefonte, PA). Mineral standards were purchased from Merck (Darmstadt, Germany). All other chemicals and reagents were of either analytical or HPLC grade and were purchased from Merck (Darmstadt, Germany).

2.2. Quality parameters of wholegrain

Hectoliter weight, thousand grain, and hardness were determined according to Cereals & Grains Association [4].

2.3. Quality parameters of flour

Zeleny sedimentation, falling number, and wet and dry gluten content were achieved by following International Association for Cereal Science and Technology [5]. Glutograph parameters were measured following the manual of Brabender [6]. Alveograph analysis was achieved by following the standard procedure of Cereals & Grains Association [7]. Gluten and gliadin were determined according to Association of Official Analytical Chemists [8].

2.4. Functional properties

Water holding capacity (WHC) and oil holding capacity (OHC)

Briefly, 1 g of the sample was mixed with 10 ml of distilled water or oil. After mixing for 30 seconds, the mixture was incubated for 30 minutes at room temperature. Then, it was centrifuged at 3000 g at 25°C for 20 minutes, and the supernatant was weighed. In Eq. (1), m_1 is the weight of the sample before mixing, and m_2 is the weight of the supernatant.

$$WHCorOHC = \frac{m_2 - m_1}{m_1}$$

1

Foaming capacity and stability

Briefly, 150 ml of distilled water was added to 4.5 grams of flour and homogenized at 9500 rpm for 2 minutes. Then, the samples were poured into a 250 ml graduated cylinder, and the foam levels were measured at 0, 5, 10, 20, 30, and 60 minutes.

The least gelling concentration

Suspensions were prepared with 5 ml of distilled water at 2–20% w/w. It was boiled in a water bath at 90°C for 1 hour. It was quickly cooled under running water and incubated at 4°C for 2 hours. The concentration at which the sample from the inverted test tube did not fall or slip was considered to be the least gelation concentration.

2.5. Bread Preparation

The formulation of the bread samples was summarized in Table S1 (Supplementary Material). The dough was kneaded in a mixer (Dioasna Laboratory Type, Germany) for 2 minutes slow and 8 minutes fast and then rested for 10 minutes. The dough was fermented for 1 hour at 38 ± 2°C and 82% RH. After fermentation, it was cooked in an oven (Wiesheu, Italy) at 230–235°C for 35 minutes.

2.6. Proximate Composition

The ash and moisture contents of the samples were determined according to the method of the Association of Official Analytical Chemists Method No. 923.03 and Method No. 925.10, respectively [9]. The total protein content was calculated by multiplying Kjeldahl nitrogen by 6.25 [9]. The crude fiber content was determined according to AOAC Method No. 962.09 [9]. Lipid content was estimated by the Soxhlet extraction method with *n*-hexane. In the Soxhlet apparatus, 1 part of the pellet was combined with 40 parts of *n*-hexane. The solvent was brought to a boiling point, and the whole oil of the samples was dissolved in *n*-hexane. The lipid fraction was extracted from the distillation flask after the pellet had been placed in the Soxhlet apparatus for 24 hours. A rotary evaporator was used to evaporate the remaining *n*-hexane. The lipid yield of the samples was estimated gravimetrically after the lipid was weighed. Carbohydrate levels were determined by subtracting the sum of the percentages of moisture, ash, protein, and lipid from 100. All measurements were carried out in triplicate.

2.7. Mineral profile

The examination for mineral assessment was realized on inductively coupled plasma mass spectrometry, ICP–MS Agilent 7700X (Agilent, Türkiye), following the International NMKL 186 instruction [10]. To determine the mineral profile, 0.2 grams of each sample was weighed and taken into vessel tubes. Briefly, 10 ml of 65% HNO_3 (Extrapure) was added and they were kept open-top for 15–20 min under a fume hood. Then, the samples were incinerated in the microwave combustion system under specified conditions. Samples were taken after microwave digestion passed through 0.45 µm syringe filters. The filtered samples were taken to the stage of determination of mineral contents in ICP-MS. Standard mineral solutions were prepared to generate a calibration curve for calculation. A 2% HNO_3 solution, prepared from 65% HNO_3 (Suprapure) with ultrapure water, was used as a blank.

2.8. Fatty acid profile

To identify and quantify each fatty acid methyl ester (FAME) in the samples, gas chromatography with a flame ionization detector (GC-FID) system was used. The fatty acid contents of the samples were analyzed using a method adapted from Uluata et al. [11]. FAMEs were analyzed using an Agilent 7820A (Agilent Technologies Inc., Palo Alto, USA) gas chromatograph equipped with a capillary column (30 m × 0.25 mm i.d., 0.25 × µm; Agilent 112–8837), a flame ionization detector (FID), and a 16-sample automatic liquid sampler. The injector and detector temperatures were maintained at 250 and 280°C, respectively. The injection volume was 1 µl. The carrier gas hydrogen flow rate was 40 mL/min, and the split ratio was 1/50. The retention times of each FAME were compared against the standard mixture of FAME to identify the FAME composition of the samples. Fatty acid composition results were expressed as a weight percentage.

2.9. Phenolic extraction

Free phenolic extraction

Whole grain samples were ground before extraction. Briefly, based on Verardo et al. [12], 0.5 grams of each sample was weighed, and 10 ml of 80% ethanol solution was added. The samples, which were vortexed for 1 minute, were kept in an ultrasonic water bath for 10 minutes. After centrifugation at 4000 rpm for 15 minutes at 10°C, the supernatant was recovered, and the residue was re-extracted. The supernatants were combined and brought up to 20 or 25 ml by volume. The prepared extracts were stored at -20°C for further analysis.

Bound phenolic extraction

Briefly, based on Verardo et al. [12], 15 ml of 2 M NaOH was added to the residue obtained in the extraction of free phenolics. After stirring for 4 hours, the pH was adjusted between 2–3 by adding the required amount of 12 M HCl. Then, 20 ml of diethyl ether: ethyl acetate solution (1:1) was added. The samples, which were kept in an ultrasonic water bath for 10 minutes, were centrifuged at 4000 rpm for 10 minutes at 10°C. The organic phase was evaporated in a rotary evaporator at 50°C. The remaining extract

was collected with 5 ml of 80% ethanol solution. The prepared extracts were stored at -20°C for further analysis.

2.10. Total flavonoid content (TFC)

The total amount of flavonoids in the extracts was determined based on Dewanto et al. [13]. Briefly, 1.25 ml of distilled water and 75 µl of 5% Na_2NO_3 solution were added to 250 µl of extract. After waiting for 6 minutes, 150 µl of 10% $AlCl_3$.6H2O was added, and after 5 minutes, 0.5 ml of 1 M NaOH was added. The total volume was brought up to 2.5 ml by adding 275 µl of distilled water, and the samples were vortexed for 10 seconds. Absorbance was read at 510 nm (BioTek Instruments, Winooski, USA), and the results were expressed in mg of rutin equivalent (RE)/100 g sample.

2.11. Total antioxidant activity (TAA)

The iron-reducing capacity of the extracts was determined based on Deighton et al. [14]. FRAP reagent was prepared by mixing 300 mM sodium acetate solution at pH = 3.6, 10 mM 2,4,6-Tris(2-pyridyl)-s-triazine (TPTZ) solution prepared with 40 mM HCl, and 20 mM $FeCl_3$ solution at a ratio of 10:1:1. Then, 900 µl of FRAP reagent was added to 100 µl of the sample, and exactly 4 minutes later, absorbance was read via a spectrophotometer (BioTek Instruments, Winooski, USA) at a wavelength of 593 nm. The results were expressed in mg Trolox equivalent (TE)/100 g sample.

2.12. Sensory analysis

To determine if there was a sensory difference between 100% modern bread (B0) and %15 Kavilca-added bread (B15), the triangular test was applied [15]. Briefly, 25 panelists aged 24–58 (mean age 33) were asked whether they noticed any difference between the samples in terms of visual, taste, odor, and texture in line with their preferences. During the experiment, the samples were coded, and the standard test procedure was applied. Taste transitions between samples were provided with water. In addition, a sensory analysis was carried out using the Quantitative Descriptive Profile (QDP) Method [16]. Ten members of the Food Engineering Department's master and doctoral students (ages 23–42) were trained to perform sensory evaluations. The descriptive test revealed the characteristics of the product in terms of flavor, appearance, texture, and smell with trained panelists. For each category, the sensory quality scores ranged from 1 to 10 according to Table S2 (Supplementary Material), presenting definitions and references for the scores.

2.13. Statistical analysis

The Minitab 18.0 licensed program was used, and the data were analyzed with one-way ANOVA and Tukey's posthoc test for multiple comparisons with statistical significance at a 95% confidence level ($p < 0.05$) to identify differences among groups. The data are given as the mean ± standard deviation.

3. Results And Discussion

3.1. Quality parameters

The grain samples differed from each other in terms of hectoliter weight and hardness. The hectoliter weight and hardness of TAG were higher than that of TDG ($p < 0.05$) (Table 1). Hectoliter weight, thousand grain, and hardness are important parameters for milling efficiency. The quality of wheat for final use is closely related to the hardness of the kernel. Hardness depends on the genetic structure of wheat as well as the amount of starch and protein. Hard wheat is more granular than soft wheat when they are milled and hard wheat is preferred for bread and pasta production. TAG was higher in hectoliter weight indicating more dry matter and flour yield. They both were classified as hard wheat.

Table 1
Quality parameters of *T. aestivum* and *T. dicoccum* whole grain and flour.

		TAG	TDG	TAF	TDF
Hectoliter weight	kg/hl	77.9 ± 0.00 a	75.3 ± 0.00 b		
Thousand grain	g	36.5 ± 0.00 a	35.5 ± 0.00 a		
Hardness (SKCS)		65.6 ± 0.01 b	73.2 ± 0.00 a		
Zeleny sedimentation	ml			42.00 ± 0.02 a	19.00 ± 0.01 b
Falling number	s			440 ± 1.01 a	351 ± 1.00 b
Wet gluten	%			28.9 ± 0.02 b	33.4 ± 0.03 a
Dry gluten	%			10 ± 0.02 b	11.2 ± 0.03 a
Gluten index	%			98.1 ± 0.02 a	32.5 ± 0.03 b
Color					
L				99.35 ± 0.03 a	97.62 ± 0.04 b
a				1.48 ± 0.01 b	1.57 ± 0.02 a
b				10.81 ± 0.03 b	12.43 ± 0.02 a
Glutograph					
Stretch	BU			525 ± 8.41 b	823 ± 10.53 a
Stretch	s			125 ± 4.30 a	5 ± 0.51 b
Relaxation	BU			417 ± 5.29 b	553 ± 6.82 a
Alveograph					
P	mm			135 ± 1.01 a	62 ± 1.05 b

TAG: *Triticum aestivum* grain. TDG: *Triticum dicoccum* grain. TAF: *Triticum aestivum* flour. TDF: *Triticum dicoccum* flour. SKCS: Single Kernel Characterization System. BU: Brabender unit. P: Maximum pressure required for the deformation of the sample or overpressure. L: Maximum amount of air the bubble is able to contain. G: Swelling index. W: Dough baking strength. Data are expressed as mean ± SD.

		TAG	TDG	TAF	TDF
L	mm			64 ± 1.02 [a]	63 ± 1.03 [a]
P/L				2.1 ± 0.01[a]	0.98 ± 0.00[b]
G				17.8 ± 0.49 [a]	14.6 ± 0.51 [b]
W	10 joule			349 ± 3.28 [a]	83 ± 1.10 [b]
Gliadin	mg/kg	92.8 ± 0.02 [a]	83.4 ± 0.03 [b]	48.3 ± 0.22 [d]	52.9 ± 0.27 [c]
Gluten	mg/kg	185.6 ± 0.59 [a]	166.8 ± 0.60 [b]	96.6 ± 0.31 [d]	105.8 ± 0.25 [c]
Phytic acid	g/100 g	0.86 ± 0.02 [b]	1.15 ± 0.04 [a]	0.86 ± 0.02 [b]	1.18 ± 0.03 [a]
Water holding capacity	g/g			1.09 ± 0.04[a]	1.06 ± 0.02[a]
Oil holding capacity	g/g			0.71 ± 0.04[a]	0.76 ± 0.07[a]
Foaming Capacity	%			34.7 ± 0.02[a]	34.7 ± 0.01[a]
Foaming Stability	%			53.8 ± 0.10[a]	50.0 ± 0.11[a]
The least gelling concentration	%			10.02 ± 0.01[b]	12.64 ± 0.02[a]

TAG: *Triticum aestivum* grain. TDG: *Triticum dicoccum* grain. TAF: *Triticum aestivum* flour. TDF: *Triticum dicoccum* flour. SKCS: Single Kernel Characterization System. BU: Brabender unit. P: Maximum pressure required for the deformation of the sample or overpressure. L: Maximum amount of air the bubble is able to contain. G: Swelling index. W: Dough baking strength. Data are expressed as mean ± SD.

The gluten amount of TDF was found to be 105.8 mg/kg (Table 1). Gluten is a wheat protein and has an effect both on human nutrition and the structural properties of bread. According to Food and Drug Administration (FDA), to label a food product as "gluten-free", it is required that the gluten level in the product offered to the end consumer does not exceed 20 mg/kg. Therefore, *T. dicoccum* is not suitable for gluten-free diets. However, a high gluten content is desired in bread production, because gluten provides elasticity to the dough via gliadin and stickiness via glutenin.

The sedimentation of TAF was approximately two times higher than that of TDF ($p < 0.05$) (Table 1). Gluten quality is an important criterion for flour quality because it traps gas bubbles during fermentation. The higher sedimentation value of TAF indicates better gluten quality. It would not be enough to use TDF alone in bread production. The use of a mixture with other wheat flour may contribute to the improvement of bread properties. The gluten index is another indicator of gluten quality. TAF has strong gluten (> 80%),

while TDF has normal gluten (30–80%) (Table 1). A higher gluten index value means better gluten quality of wheat.

The relaxation (BU) value obtained from the glutography is evaluated as the measure of the elasticity of the dough, and the stretch (s) is the measure of the elongation of the dough. TAF (125 s) was found to be more stretched than TDF (5 s) ($p < 0.05$). As gluten strength and quality increase, relaxation value decreases, and stretch (s) value increases. The gluten quality of TDF was lower when compared with the different bread wheat cultivars that belonged to Regional Yield Trials (Central Research Institute for Field Crops, Ankara, Türkiye) [17]. Although TDF had a significantly higher amount of gluten and gliadin than TAF ($p < 0.05$), the gluten quality of TDF was lower than TAF.

Another important quality criterion of flour is α-amylase activity measured by falling number. Falling number (FN) analysis revealed that the drop time of TAF was longer than TDF ($p < 0.05$) (Table 1). α-amylase breaks down starch and lower starch causes lower viscosity of the paste; therefore, a higher falling number indicates lower α-amylase activity. TAF had a higher FN than TDF, but both were appropriate for bread making, which was > 250 s [18].

The principle of alveograph analysis, which gives information about the viscoelastic structure of the dough, is based on keeping the dough formed under certain conditions for a while and measuring its resistance against swelling by blowing air into it. Alveograph parameters are pressure (P), length (L), energy (W), and swelling index (G). P represents the maximum pressure during the inflation of the dough. As seen in Table 1, TAF (135 mm) had a significantly higher P value than TDF (62 mm) ($p < 0.05$). A high P value indicates stronger gluten which is desired in bread production. L refers to the extensibility of the dough and there was no significant difference between the L values between TAF and TDF ($p > 0.05$). W is the amount of energy required to inflate the dough until it bursts. It refers to the amount of CO_2 gas released by microorganisms during fermentation and it has a significant effect on the volume and texture of bread in bread making. TDF had significantly lower W than TAF which would lead to not swollen bread as much as modern bread ($p < 0.05$).

There was no significant difference between TAF and TDF in terms of water holding, oil holding, and foam stability and capacity ($p > 0.05$). TAF and TDF were able to absorb more water than their weight. The least gelation concentration of TDF (12.64%) was significantly higher than that of TAF (10.02%) ($p < 0.05$) (Table 1). Water holding capacity is related to amylopectin and amylose content since the amorphous structure of starch is responsible for hydrogen binding during interaction with water. Oil holding capacity is related to the physical entrapment of oil to the apolar chain of the protein. Higher oil holding capacity better mouthfeel and longer flavor retention. It also indicates the lipophilic nature of flavor constituents [19]. Foaming capacity is related to the configuration of protein molecules and small air bubbles are trapped during foaming. Foaming capacity and stability are important quality parameters since they indicate how much CO_2 could be trapped in the flour matrix during fermentation which would lead higher or lower volume of bread [19]. Gel formation during heating is preferred in numerous bakery products and gelling concentration is related to starch and protein content [20]. According to the results,

although there was a minimal difference, TDF owned less ability to form gel during heating than TAF ($p < 0.05$). The lower protein content of TDF might have caused lower gelation ability since low protein and high insoluble fiber hamper flour from swelling and generate a multi-dimensional network during heating [20].

3.2. Proximate composition

The protein content of wholegrains, flour, and bread samples varied between 8.35–14.28% (Table S2, Supplementary Material). There was no significant difference in protein content in *T. aestivum* and *T. dicoccum* wheat and flours ($p > 0.05$). Fat content varied between 1.10–3.10%, while crude fiber is between 5.39–22.25%.

3.3. Mineral profile

Mineral deficiency adversely affects human health, especially in countries where there is insufficient consumption of fruits, vegetables, and foods of animal origin, and where basic foods with low microelement concentrations, namely cereals, are consumed more and diet diversity is low. In addition, it is an important issue for human health to monitor how much toxic elements are taken into the body to ensure food safety. Since wheat contributes significantly to the daily calorie requirement, *T. aestivum* and *T. dicoccum* and wheat samples were investigated in terms of calcium (Ca), magnesium (Mg), phosphorus (P), sulfur (S), iron (Fe), zinc (Zn), and aluminum (Al) (Table 2). There was no significant difference between the macroelement and microelement contents of the samples ($p > 0.05$). However, TDG and TDF (2.85 mg/kg dw and 5.16 mg/kg dw), contained approximately 7 times less Al than TAG and TAF (27.98 mg/kg dw and 26.89 mg/kg dw) ($p < 0.05$).

Table 2
Mineral profile of *T. aestivum* and *T. dicoccum* whole grain and flour.

Minerals		TAG	TDG	TAF	TDF
Macro elements (mg/100g dw)	Ca	34.30 ± 10.45[a]	22.35 ± 10.65[a]	34.45 ± 10.46[a]	26.44 ± 10.71[a]
	Mg	149.80 ± 54.47[a]	151.89 ± 55.21[a]	124.00 ± 45.09[a]	156.27 ± 57.03[a]
	P	417.29 ± 125.97[a]	463.51 ± 143.78[a]	323.93 ± 100.48[a]	468.25 ± 145.25[a]
	S	190.26 ± 38.05[a]	182.45 ± 36.49[a]	168.05 ± 33.61[a]	172.08 ± 34.42[a]
Essential elements (mg/kg dw)	Fe	59.15 ± 22.99[a]	26.57 ± 10.33[a]	52.48 ± 20.40[a]	35.13 ± 13.65[a]
	Zn	11.96 ± 4.64[a]	24.80 ± 9.63[a]	13.47 ± 5.23[a]	31.20 ± 12.11[a]
Non-essential elements (mg/kg dw)	Al	27.98 ± 5.60[a]	2.85 ± 0.68[b]	26.89 ± 5.38[a]	5.16 ± 1.15[b]

TAG: *Triticum aestivum* grain. TDG: *Triticum dicoccum* grain. TAF: *Triticum aestivum* flour. TDF: *Triticum dicoccum* flour. Data are expressed as mean ± SD.

The factors that affect the mineral content of wheat are mainly the environment, genotype, growing conditions, and soil composition [21]. In a study on the mineral profiles of *T. dicoccum* subspecies, it was stated that wild wheat contains large amounts of Mg among macroelements, and Mn, Fe, and Zn contents among microelements [10]. Additionally, Del Coco et al. [10] emphasized that less toxic elements accumulate in wild wheat compared to modern wheat, especially in terms of cadmium. Al is particularly common in *T. aestivum* but is a toxic element that is commonly found in acidic soil conditions that limit crop production [21]. Also, Silva et al. [21] stated that Al interacts negatively with P, Mg, and K in wheat. Physical processes such as milling reduce the mineral content while improving the usability of wheat due to the reduction in antinutrient content. Dietary exposure to aluminum from wheat flour is an important health issue. de Paiva et al. [22] mentioned that in their study daily intake of three portions of cereal-based baby food contributes up to 10.48% of the provisional tolerable weekly intake of aluminum which was determined as 2 mg/kg body weight by the Joint FAO/WHO Expert Committee on Food Additives (JECFA). Although that ratio carries no risk, Guo et al. [23] stated that especially children were at a higher risk of excess aluminum exposure from wheat flour in Shangai, China. Therefore, integrating the *T. dicoccum* (Kavilca) into the formulation of cereal-based baby food might be a better choice to decrease as possible the accumulation of toxic elements.

3.4. Fatty acid profile

The amount of lipids in wheat grain is quite low (between 3 and 4%), and they are mostly found in the germ [24]. Most of the fatty acids found in wheat are unsaturated (C18:1, C18:2, and C18:3) and two of them are essential (linoleic and linolenic) [25]. Some of the fatty acids included in the wheat grain such as palmitic, oleic, and linoleic acids are crucial for human health. Essential fatty acids are building blocks of several biomolecules in the human body (like membrane phospholipids) and are involved in metabolic activities like regulating blood lipid levels, particularly cholesterol [25].

The fatty acid profile significantly affects the technological properties of wheat, and the prevalence of unsaturated fatty acids in wheat is indicative of the nutraceutical effects of food. The fatty acid profiles of the samples are given in Table 3. *T. aestivum* and *T. dicoccum* were found to be rich in linoleic acid, ranging from 51.20–60.20%. TAG and TAF (60.20% and 59.10%) had higher linoleic acid contents than TDG and TDF (51.20% and 51.30%) ($p < 0.05$). Oleic acid was the second dominant fatty acid in the samples. TDG and TDF contained significantly higher amounts of oleic acid than TAG and TAF. In addition, the highest amount of palmitic acid was found in TDF (17.10%) ($p < 0.05$). Oleic and linoleic acid contents were determined to be the most essential fatty acids in the flour of *T. aestivum* and *T. durum* [24]. In the literature studies, linoleic, palmitic, and oleic acid content have a higher abundance among the other fatty acids in flour samples [24]. The findings of this study are compatible with the literature according to this aspect. On the other hand, the abundance of the double bond in the fatty acids is responsible for peroxidation and rancidification in the food.

Table 3
Fatty acid profiles of *T. aestivum* and *T. dicoccum* whole grain and flour.

Fatty acid profile		%			
		TAG	TDG	TAF	TDF
SFA					
C 16:0	Palmitic acid	16.60 ± 1.80 [a]	16.40 ± 1.73 [a]	16.50 ± 1.56 [a]	17.10 ± 1.31 [a]
C 18:0	Stearic acid	1.10 ± 0.21 [ab]	1.50 ± 0.25 [a]	0.90 ± 0.16 [b]	1.60 ± 0.23 [a]
C 20:0	Arachidic acid	4.00 ± 0.33 [a]	3.90 ± 0.29 [a]	4.10 ± 0.22 [a]	3.60 ± 0.24 [a]
	Total (%)	21.7	21.8	21.5	22.3
MUFA					
C 18:1 (cis)	Oleic acid	18.10 ± 1.46 [b]	25.20 ± 1.72 [a]	17.20 ± 1.55 [b]	24.80 ± 1.61 [a]
	Total (%)	18.10	25.20	17.20	24.80
PUFA					
C 18:2	Linoleic acid	59.10 ± 2.01 [a]	51.30 ± 1.85 [b]	60.20 ± 2.17 [a]	51.20 ± 1.93 [b]
C 18:3	Linolenic acid	1.10 ± 0.15 [ab]	1.40 ± 0.17 [a]	0.80 ± 0.11 [b]	1.30 ± 0.15 [a]
C 18:3 n6	γ-Linolenic acid	ND	0.30 ± 0.01 [a]	0.20 ± 0.01 [b]	0.30 ± 0.02 [a]
	Total (%)	60.2	53.00	61.20	52.80

TAG: *Triticum aestivum* grain. TDG: *Triticum dicoccum* grain. TAF: *Triticum aestivum* flour. TDF: *Triticum dicoccum* flour. Data are expressed as mean ± SD. ND: Not detected.

TDG and TDF had a higher amount of stearic acid, linolenic acid, and γ-linolenic acid ($p < 0.05$). There may be differences between the fatty acid profiles of the harvests of the same species in different periods and geographical origins [26]. Other biotic and abiotic stresses that affect the fatty acid profile of wheat species are growth temperature, humidity, rainfall level, salt, drought, pathogens, and others. For instance, compared to soft white wheat, durum wheat and hard red wheat typically have a higher amount of lipid, and the quantities of fatty acids vary between the two types of wheat. The necessity for membrane fluidification during cold weather encourages growth in lipid content in wheat and a larger degree of unsaturation in fatty acids is observed in this climate [25].

3.5. Total flavonoid content (TFC) and total antioxidant activity (TAA)

Phenolic acids and flavonoids are the most prevalent phenolic substances found in grains. Flavonoids in grains present as glycosides joined to other sugar molecules or in combinations with other phenols,

organic acids, amines, lipids, or carbohydrates rather than aglycones and methylated derivatives. They are primarily localized in the aleurone layer, seed coats, and embryos in either free or bound form [27]. The TDF (3.65 mg RE/g dw) had a higher TFC than the TAF (Table 4). This result supported that ancient wheat has higher phenolic acids and total polyphenol content [28]. This difference in the composition of TDF and TAF may be due to basically environment and genotype [3]. In other studies, Sharbati variety of *T. aestivum* had 0.365 mg QE/g and Heibaoshi variety had 0.319 mg RE/g while in this study, TAF (Esperia variety of *T. aestivum*) revealed 3.11 mg RE/g dw (wet weighed: 3.53 mg RE/g), approximately 10-fold higher [29, 30]. The high TFC of the TAF might be explained by how the phenolic content is affected by genotype, environmental variations, and cropping years [3]. From a different perspective, TDF had a higher TFC than TDG, significantly, indicating that finer products revealed a higher TFC. This positive impact could be related to finer particles having a higher surface area [31].

Table 4
Total flavonoid content and antioxidant activities of wholegrains and flour of *Triticum aestivum* and *Triticum dicoccum*.

		TAG	TDG	TAF	TDF
Total Flavonoid Content (mg RE/g DW)	Free	1.82 ± 0.36 ^{a}	1.97 ± 0.33 ^{a}	1.84 ± 0.11 ^{a}	1.95 ± 0.21 ^{a}
	Bound	1.62 ± 0.23 ^{a}	1.41 ± 0.23 ^{a}	1.27 ± 0.43 ^{a}	1.69 ± 0.28 ^{a}
	Total	3.44 ± 0.24 ^{ab}	3.38 ± 0.11 ^{ab}	3.11 ± 0.45 ^{b}	3.65 ± 0.16 ^{a}
Total Antioxidant Activity (FRAP) (mg TE/g DW)	Free	0.57 ± 0.04 ^{a}	0.58 ± 0.03 ^{a}	0.66 ± 0.02 ^{a}	0.66 ± 0.10 ^{a}
	Bound	1.67 ± 0.23 ^{a}	1.36 ± 0.12 ^{a}	1.59 ± 0.18 ^{a}	1.31 ± 0.29 ^{a}
	Total	2.25 ± 0.21 ^{a}	1.94 ± 0.14 ^{a}	2.25 ± 0.20 ^{a}	1.98 ± 0.16 ^{a}

TAG: *Triticum aestivum* grain. TDG: *Triticum dicoccum* grain. TAF: *Triticum aestivum* flour. TDF: *Triticum dicoccum* flour. RE: Rutin Equivalent. TE: Trolox Equivalent. Data were represented as mean values ± SD; a-b: Different superscript letters in the same row were significantly different ($p < 0.05$).

Antioxidant activity is greatly influenced by the presence, arrangement, structure, and total quantity of sugar moieties in flavonoid glycosides. In this study, there was no significant difference between *T. aestivum* and *T. dicoccum* in terms of antioxidant activity ($p > 0.05$). The TDF which was *T. dicoccum* grown in Kars, Türkiye had approximately 2.5 times higher TAA than *T. dicoccum turgidum* having 0.68–0.75 mg TE/g dw antioxidant activity cultivated in Italy [28]. This result might be related once more that environment and genotype affect phenolic composition [3]. Although flavonoid glycosides often exhibit lower antioxidant potency than aglycones, in the present study, bound phenolics (1.31–1.67 mg TE/g dw) had greater antioxidant activity than free phenolics (0.57–0.66 mg TE/g dw) (Table 4). This result was expected since wheat generally has more insoluble bound flavonoid glycosides than free aglycones, specifically C-linked glycosides which are linked to sugars at the C-8 or C-6 position [27]. Furthermore, phenolic acids such as ferulic acid, p-hydroxybenzoic acid, vanillic acid, syringic acid, syringaldehyde,

and p-coumaric acid were found in higher amounts in insoluble bound phenolic extracts than soluble free extracts [28].

3.6. Sensory analysis

Sensory evaluation has an important role in developing a new food product, as it directly reflects what the consumer prefers and dislikes in terms of appearance, smell, texture, aroma, and taste. The views of the bread samples are presented in Fig. 1. As a result of the triangle test, 17 of the 25 panelists participating in the experiment were able to correctly select a different bread sample among the three samples presented. In the standard experimental procedure, it was stated that the correct selection of 15 or more participants is required for $\alpha = 0.01$ in a sensory analysis using the 25-person triangular test [15]. According to this result, it can be said that there was a strong difference with 99.9% probability between the B0 and B15 samples.

For the descriptive test, bread samples were evaluated in terms of the properties defined in Table S2 (Supplementary Material). Spider diagrams of the descriptive test were presented in Fig. 2. In terms of appearance, it was determined that B100 had a darker crust and crumb color than B0 and B15 and smaller internal pore sizes ($p < 0.05$). In addition, B100 was found to be less swelled than B0 and B15 and received less like than B0 and B15 in terms of general appearance (Table 3.11) ($p < 0.05$). When evaluated in terms of textural properties, it was concluded that B100 was moister and has a firmer texture than B0 and B15 ($p < 0.05$). In terms of flavor and aroma properties, B100 was found to have a more sourish, grainier, and more persistent taste than B0 and B15 ($p < 0.05$). However, there was no statistical difference between bread samples in terms of general odor, general texture, and general taste and aroma ($p > 0.05$). Finally, there was no statistical difference between the bread in terms of overall liking ($p > 0.05$).

The findings of the triangle test indicated that the addition of less than 15% *T. dicocccum* was needed to overcome the perception of detectable sensory differences between modern bread and ancient wheat-added bread samples. These detectable differences might be swelling of the bread and moistness of the crumb since the descriptive test revealed that B15 significantly differed from B0 in terms of swelling and crumb moisture (Table S4, Supplementary Material). However, descriptive test results indicated that although there were some significant differences in terms of various aspects of the appearance, texture, and taste both 15% added and 100% *T. dicoccum* bread was found to be acceptable and overall liked by the panelists.

4. Conclusion

T. dicoccum, “Kavilca”, Emmer-type Anatolian wheat, was characterized in terms of quality parameters, functional properties, mineral, and fatty acid profile, total flavonoid, and total antioxidant activity. Novel findings of this study were low gluten quality, low Al content, and high TFC content of Kavilca along with a similar overall liking sensory score with modern bread. Although the gluten quality of *T. dicoccum* flour was lower than that of *T. aestivum*, it was acceptable for bread production. There was no significant difference between the water holding, oil holding, and foaming capacity of *T. dicoccum* and *T. aestivum*

flour. *T. dicoccum* had 7 times less toxic element Al which is an important health issue, especially for the daily Al intake of children from wheat. Among fatty acids, *T. dicoccum* was remarkable in terms of oleic acid. Moreover, although there was no significant difference between *T. dicoccum* and *T. aestivum* in terms of total antioxidant activity, *T. dicoccum* flour had the highest total flavonoid content. Furthermore, the triangle sensory test revealed that the addition of less than 15% *T. dicoccum* flour was needed to cover the sensory differences of modern bread. Overall, 15% and 100% Kavilca bread was liked and accepted by the panelists. The strength of the study is that Kavilca was characterized holistically, including quality, mineral, fatty acid, total flavonoid, and antioxidant activity. In addition, sensory analysis results might be a reference for the food industry to integrate Kavilca into several bakery products.

Declarations

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Ethical statement

The authors declare that the appropriate protocols for protecting the rights and privacy of all participants were utilized during the execution of the research, e.g. no coercion to participate, full disclosure of study requirements and risks, written or verbal consent of participants, no release of participant data without their knowledge, ability to withdraw from the study at any time.

Competing interest

I declare that the authors have no competing interests as defined by Springer, or other interests that might be perceived to influence the results and/or discussion reported in this paper.

Consent for publication

Informed consent was obtained from all individual participants included in the study.

Authors' contributions

E.F.A wrote the main manuscript text. E.F.A, E.D., Z.M., H.Y., A.S., B.Ç. realized the analyzes. B.Ö. administered the study and reviewed the manuscript.

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Availability of data and materials

The authors confirm that the data supporting the findings of this study are available within the article [and/or] its supplementary materials.

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Figures

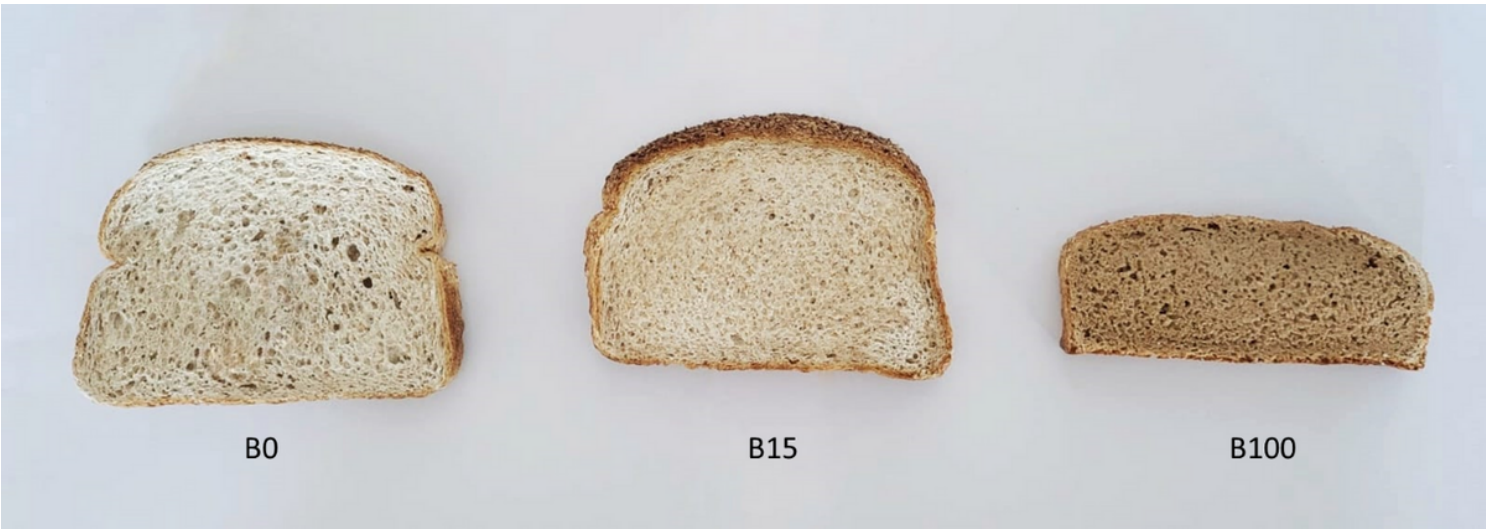


Figure 1

The visual appearance of bread samples

Bread with 100% *Triticum aestivum* flour (B0), Bread with 15% *Triticum dicoccum* flour (B15), and bread with 100% *Triticum dicoccum* flour (B100).

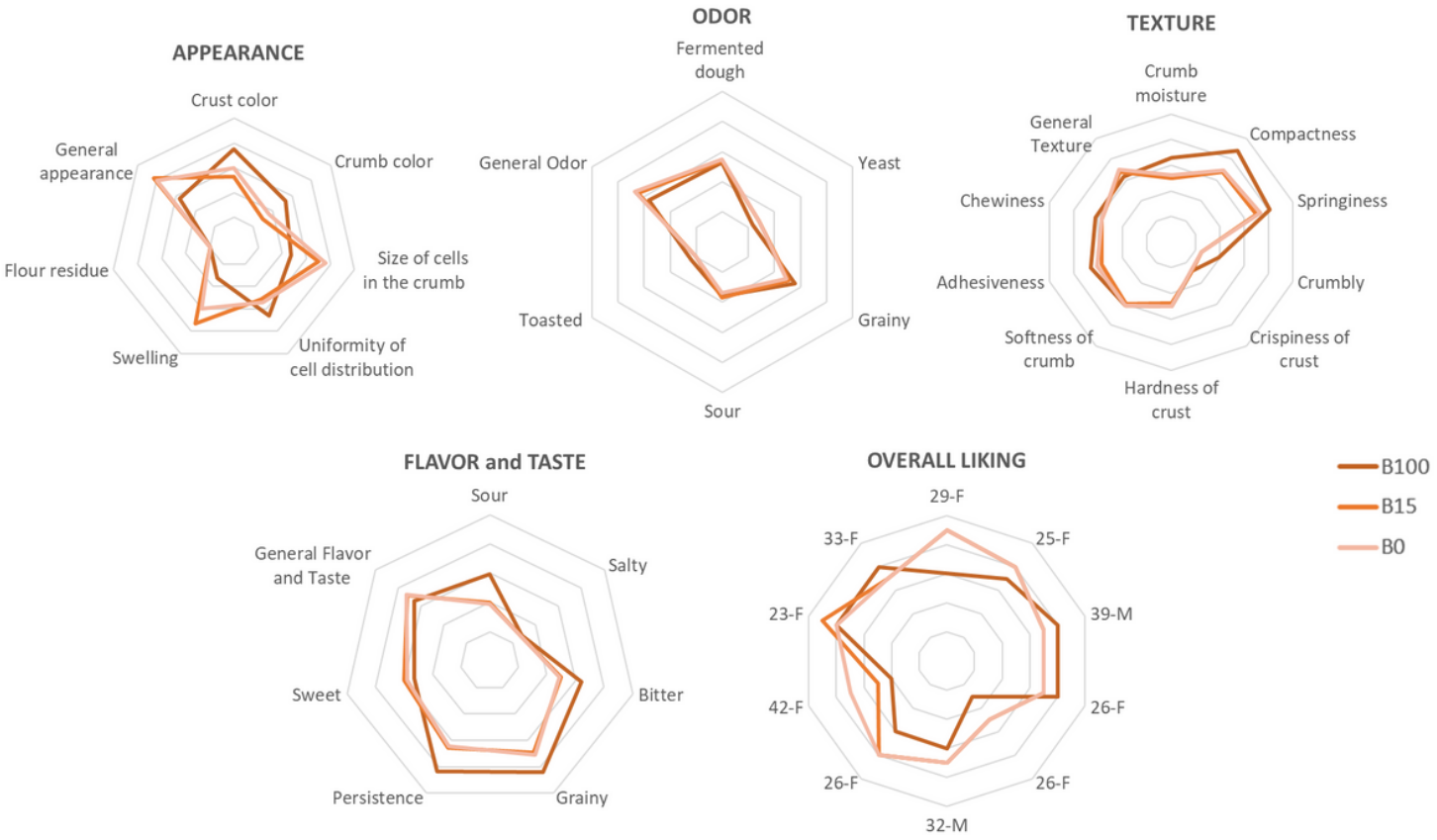


Figure 2

Spider diagrams of bread samples in terms of appearance, odor, texture, flavor, taste, and overall liking

Bread with 100% *Triticum aestivum* flour (B0), Bread with 15% *Triticum dicoccum* flour (B15), and bread with 100% *Triticum dicoccum* flour (B100). In the overall liking diagram the number indicates age of the participant, F: Female, M: Male.

Supplementary Files

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