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EFFECT OF QUINOA (*Chenepodium quinoa* WILD) FLOUR ADDITION ON THE NUTRITIONAL AND CHEMICAL PROPERTIES OF BREAD

Berat Demir[1,*], Mehmet Sahin[1], Aysun Gocmen Akcacik[1], Seydi Aydogan[1], Sumeyra Hamzaoglu[1] and Mustafa Kursat Demir[2]

[1]Bahri Dagdas International Agricultural Research Institue, Karatay, Konya, Turkey; [2]Department of Food Engineering, Engineering and Architecture Faculty, Necmettin Erbakan University, Koycegiz Campus, 42050, Konya, Turkey.
[*]Corresponding author's e-mail: berat.demir@tarimorman.gov.tr

In recent years, the negative changes created by climate change on plants have directed people to search for different plants. Some plants are drought-tolerant, and the water demand is less than other plants. Quinoa (*Chenepodium quinoa* Wild.) is a gluten-free, single-year plant that contains more protein and low carbohydrates than wheat. It has well-balanced amino acid composition when compared with reference samples from the WHO. Also, quinoa can tolerate various abiotic stresses such as different soil pH, salinity, high solar radiation, cold and night frost. In this study, properties of bread made from quinoa and wheat flour were investigated. Quinoa flour in different ratios (0, 5, 10, 15 and 20 %) was replaced by wheat flour. According the results, Phytic acid content of bread samples increased by 43.6% and TPC content increased by 11.4% with the addition of maximum (20%) quinoa flour. Also, Ca, K, Fe, Mg, Mn and Zn contents of bread samples increased by 27.1, 68.1, 21.2, 57.30, 74 and 11.2 %, respectively with the addition of maximum (20%) quinoa flour.
Keywords: Food security, bread, nutrition, phenolic, phytic acid, quinoa.

INTRODUCTION

The factors such as rapid increase in the world population, contamination of natural resources and lack of education increased the nutrition problems and complicated the reliable food supply. Quinoa (*Chenepodium quinoa* Wild.) is a drought resistant plant which has high protein and fiber ratio and low carbohydrate content according to wheat (Geerts *et al.*, 2009; Oelke *et al.*, 1992; Valencia-Chamorro, 2003). It belongs to *Chenopodiaceae* family and double-dicothyledon (Miranda *et al.*, 2012). Quinoa, which is mainly grown in the highland of Bolivia and Peru, and it contains large amounts of lipids, minerals, and high quality protein, which include essential amino acids, such as histidine and lysine (Ranhotra *et al.*, 1993; Chauan *et al.*, 1992). Although it is not known exactly when quinoa cultivation began on earth, it is estimated that it has been cultivated by the Native Americans of Central and South America since 3000 B.C. Quinoa, the plant of the Andes in South America, is the main nutrient of Aztec and Incas from ancient civilizations in this region (Tan and Yöndem, 2013). Quinoa adapts to unfavorable climatic conditions. It can grow at temperatures between -4°C and 38°C and relative humidity from 40 to 88% (FAO, 2011). It is resistant to lack of the soil moisture. Even at total annual precipitation of 100-200 mm, quinoa provides acceptable yields (FAO, 2011; Jacobsen *et al*., 2003).

Quinoa, which draws attention with its nutritional superiority, is known to have some anti-nutritional properties. Especially saponins and phytic acid are some of the anti-nutritional factors (Valencia-Chamorro, 2003). Anti-nutritional factors such as phytic acid reduce the glycemic index in small intestines by effecting the digestibility of starch (Thompson *et al*., 1987). The studies have shown that phytic acid may have beneficial effects on diet (Şat and Keles, 2004). In rats, studies have shown that phytic acid decreases blood lipids, decreases blood glucose response and cancer risk and has positive effects on health. It is also recommended to use high phytate diet against antidote activity in the prevention of dental caries, platelet aggregation, hypercalciuria treatment, nutrition of people with kidney stones and acute lead poisoning (Graf and Eaton, 1984; Forbes *et al*., 1984; Shi *et al*., 2004). Quinoa is rich in some phenolic compounds. Gawlik-Dziki *et al.* (2013) reported that quinoa leaves may inhibit many cancers, particularly prostate cancer, because they have high bioactive components (especially ferulic, sinapinic and gallic acids, kaempferol, isorhamnetin, and routine). Quinoa is a good source of some minerals which are essential for human nutrition. It contains more iron (Fe), calcium (Ca), magnesium (Mg) and zinc (Zn) than common cereals, and the iron content is also quite high (Jancurova *et al*., 2009).

Bread is the most important source of carbohydrates and widely used in grain-based nutrition in the world. It is obtained by mixing wheat flour, yeast, salt and a certain amount of water kneading dough, fermenting for a certain time and cooking. It has neutral aroma, it is cheap and easily available (Elgün and Ertugay, 1995). It is also advantageous for consumers to improve their functional and nutritional properties by enriching the bread wheat flour with quinoa flour with nutritional superiorities.

Wheat has an important place in human nutrition. One of the basic ingredients of human nutrition is bread. It is the oldest known food ingredient of mankind. Wheat quality directly affects the quality of bread. For this reason, flours which belong to different wheat varieties are mixed for to find the most suitable usage in flour industry. This investigation was undertaken to produce a high nutritional and chemical characteristics food product using quinoa flour. Therefore, this study aimed to investigate the production of bread which was produced with different wheat variety flours with different ratios of quinoa flour and determine the chemical and nutritional properties of bread samples.

MATERIALS AND METHODS

Wheat samples were tempered 14.5% moisture content according to AACC 26-95 method. Then, these samples were milled to 65 ± 1% "wheat flour" extraction rate on a lab-roller flour mill with 70 GG sieve (Brabender Junior mill) according to AACC 26-50 method. Quinoa seeds were obtained from Bora Agricultural Products, İstanbul, Turkey. The quinoa seeds were reduced to a certain size by using a sieve system with 500 µ mesh in Retsch ZM 200 laboratory mill. For the purpose of size control, all ground quinoa seeds were sieved with a 500 µ sieve and used as quinoa flour in trials. To prepare the flour blends, the fine ground "quinoa flour" and "wheat flour" were mixed thoroughly. Thus, different flour samples were obtained.

***Experimental design*:** Four different wheat samples (Tosunbey, Bezostaja-1, Karahan-99 and Konya 2002) and five different quinoa flour rates (0, 5, 10, 15 and 20%) were used as factors with two replications in the experiments, according to the completely randomized design of (4 × 5) × 2 factorial plans.

***Bread-making process*:** Bread samples were obtained by the modification of AACC method 10-10B (AACC 2000). Fermentation time was "30+30" min. for bulk fermentation and was 55 min. for proofing. Then, the samples were dried at 50°C for 18 h in an air-convection oven (Nuve FN-500, Ankara, Turkey) and these samples were crushed using a laboratory type grinder (Moulinex Super Junior S, Paris, France). Afterwards the samples were packed in sealed bags and stored at 5°C, until used.

Chemical and Nutritional Analyses:

***Total phenolic content (TPC)*:** The TPC of wheat flour, quinoa and bread samples were evaluated as colorimetric using a modified version of the Folin–Ciocalteu assay as described by Slinkard and Singelton (1977) and Gamez-Meza *et al*. (1999). All samples (4 g) were shaken in acidified methanol (HCl/methanol/water, 1:80:10, v/v) for 2 hours in a shaking water bath (24 ± 1°C), then the mixture was centrifuged at 3000 rpm (Sigma 2-16, Germany) for 10 minutes. The TPC was determined after centrifugation by using the obtained supernatant (Gao *et al.*, 2002; Beta *et al*., 2005). 0.1 ml supernatant, 0.5 ml Folin-Ciocaltaeu reagent (10%, v/v, water) and 1.5 ml sodium carbonate solution (20%, w/v, water) were mixed in the test tube for 2 hours at room temperature (Incubated at 24 ± 1°C). At the end of this period, the absorbance values of the solutions were read at 760 nm wavelength spectrophotometer (Hitachi-U1800, Japan) and the TPC was calculated as gallic acid equivalent.

***Phytic acid*:** Phytic acid was measured by a colorimetric method according to Haug and Lantzsch (1983). Phytic acid in the sample was extracted with a solution of HCl (0.2 N) and precipitated with a solution of Fe III (ammonium iron (III) sulphate 12 H_2O). The Fe amount in the serum part was measured with a spectrophotometer and phytic acid amount was calculated accordingly.

***Mineral content*:** The mineral (Ca, Fe, Mg, Zn, K and Mn) contents of the samples were determined by inductively coupled plasma atomic emission spectrometry (ICP-AES) (Vista series, Varian International AG, Switzerland) with an automatic sampler system. Approximately 0.5 g of the sample was put into a burning cup, and 5 mL of HNO_3 + 5 mL H_2SO_4 was added. The samples were incinerated in a microwave oven (Mars 5, CEM Corporation, USA). The solution was diluted to 100 mL with water. Concentrations were determined by ICP-AES (Bubert and Hagenah, 1987).

***Statistical analyzes*:** The trials were carried out according to factorial trial design in randomized plots of 4 x 5 x 2 with 2 replications (Duzgüneş *et al.*, 1987). At the end of the research, statistical analyzes were done by using JMP11 (Anonymous, 2014) program.

RESULTS AND DISCUSSION

Analytical results: The results for the analysis of the raw materials (quinoa flour and wheat flour samples) used in the production of bread samples are presented in Table 1. Phytic acid, TPC and some mineral content of quinoa flour were found higher than those of the wheat flours. Phytic acid content of quinoa flour was determined 988.91 mg/100g and TPC of quinoa flour as 1.79 mg GAE/g. According to Ruales and Nair (1993) the content of phytic acid in quinoa seeds is about 1% of the dry matter. Koziol (1992) determined phytic acid amount between 1050 and1350 mg/100g in quinoa. Phytic acid amount in bread wheat varieties ranged 350.12-541.11mg/100g. The highest phytic acid value was recorded in Tosunbey while the lowest value was observed in Konya 2002 and Karahan-99 variety flour.

TPC of bread wheat varieties ranged from 0.74 to 1.11 mg GAE/g and TPC of quinoa was determined as 1.79 mg GAE/g. Jan *et al.* (2016) found 2.41 mg/g TPC of quinoa in a study. Among wheat flours, the highest TPC value was

Table 1. Phytic acid, TPC and mineral content of raw materialas.

Variety	Phytic acid (mg/100g)	TPC (mg GAE/g)	Minerals (mg/100g)					
			Ca	K	Fe	Mg	Mn	Zn
Konya-2002	350.12^{d**}	1.05^{d**}	16.44^{b**}	127.02^{e**}	1.25^{b*}	36.45^{b**}	0.20^{b*}	1.92^{b*}
Bezostaja-1	456.89^{c}	1.11^{c}	14.32^{c}	133.30^{c}	1.44^{b}	33.00^{c}	0.20^{b}	1.83^{b}
Tosunbey	541.11^{b}	0.74^{e}	14.01^{c}	143.09^{b}	1.51^{b}	33.60^{c}	0.28^{b}	1.70^{b}
Karahan-99	378.25^{d}	0.77^{b}	14.93^{c}	130.54^{d}	1.22^{b}	31.78^{d}	0.23^{b}	0.99^{c}
Quinoa Flour	988.91^{a}	1.79^{a}	40.04^{a}	674.03^{a}	3.40^{a}	154.60^{a}	1.25^{a}	3.42^{a}

**: $p < 0.01$; *: $p < 0.05$; The means with the same letter in column are not significantly different ($p<0.05$), TPC: Total phenolic content.

obtained from Bezostaja-1 and the lowest value was obtained from Tosunbey wheat flour.

Also, Ca, K, Fe, Mg, Mn and Zn content of quinoa flour was determined as 40.04, 674.03, 3.4, 154.6, 1.25 and 3.42 mg/100g, respectively (Table 1). Konishi *et al*. (2004) studied the distribution of minerals in quinoa grain. They found that phosphorus (P), magnesium (Mg) and potassium (K) were localized especially in the embryo. K and Ca were present in the pericarp, may be associated with pectin. In another study, Sanders (2009) determined Ca, Fe, K, Mg and Zn content of quinoa as 56.5, 1.4, 1193, 176 and 2.8 mg/100g, respectively. This was an expected because quinoa is a very rich source of minerals. It contains more calcium, magnesium, iron, and zinc than common cereals, and the iron content is particularly high (Jancurová *et al.*, 2009). According to the results of variance analysis, the variety interaction of blends on Ca, K, Mg, TPC and phytic acid values were significant at $p < 0.01$; Fe, Mn and Zn values were significant at $p < 0.05$ level (Table 1).

***Bread analyzes*:** Some chemical and nutritional properties of bread samples are given in Table 2 and 3. According to the results of variance analyzes, wheat variety and quinoa flour ratio factors were found significant at $p <0.01$ level regarding amount of TPC and phytic acid content in bread samples. When the results were evaluated in terms of quinoa flour ratio, it was observed that the amount of TPC and phytic acid content in bread samples increased statistically as the rate of quinoa flour increased.

Table 2. Phytic acid content and TPC of bread materials.

Varieties	Phytic acid (mg/100g)	TPC (mg GAE/g)
Bezostaja-1	115.92^{a**}	1.10^{a**}
Karahan-99	91.56^{c}	0.77^{c}
Konya-2002	106.01^{b}	1.03^{b}
Tosunbey	72.03^{d}	0.76^{c}
Quinoa Flour (%)		
0	79.24^{e**}	0.87^{d**}
5	87.84^{d}	0.90^{c}
10	95.82^{c}	0.91^{c}
15	105.15^{b}	0.94^{b}
20	113.85^{a}	0.97^{a}
Increase by maximum (20%) quinoa flour replacement (%)	43.60	11.40

**: $p<0.01$; The means with the same letter in column are not significantly different ($p<0.05$). TPC: Total phenolic content.

There were significant changes in total mineral contents (TMC) of breads containing different ratios of quinoa flour (Table 3). According to the results of variance analysis, wheat variety factor was found significant at $p <0.01$ level for Ca, K, Fe, Mg and Zn content and nonsignificant on the Mn content in bread samples. In addition, quinoa flour ratio factor was found to be significant at $p <0.01$ level on the Ca, K, Mg, Mn and Zn content and significant at $p <0.05$ level on Fe content in bread samples. The replacement of wheat flour with quinoa flour and increasing the ratios of this replacement

Table 3. Mineral content of bread materials (mg/100g).

Wheat variety	Ca	K	Fe	Mg	Mn	Zn
Bezostaja-1	20.28^{c**}	209.81^{b**}	1.86^{ab**}	47.96^{c**}	$0.39a^{ns}$	2.04^{ab**}
Karahan-99	23.78^{a}	208.68^{c}	1.97^{ab}	45.89^{d}	0.38^{a}	1.41^{c}
Konya-2002	21.36^{b}	208.16^{c}	1.78^{b}	51.95^{a}	0.36^{a}	2.12^{a}
Tosunbey	20.17^{c}	212.78^{a}	2.31^{a}	48.67^{b}	0.36^{a}	1.91^{ab}
Quinoa flour rate (%)						
0	18.63^{e**}	156.22^{e**}	1.79^{b*}	37.87^{e**}	0.27^{c**}	1.77^{c**}
5	20.49^{d}	182.76^{d}	1.90^{ab}	42.95^{d}	0.32^{bc}	1.80^{c}
10	21.49^{c}	209.42^{c}	1.97^{ab}	48.54^{c}	0.39^{abc}	1.87^{b}
15	22.71^{b}	238.25^{b}	2.08^{ab}	54.18^{b}	0.43^{ab}	1.93^{ab}
20	23.68^{a}	262.64^{a}	2.17^{a}	59.57^{a}	0.47^{a}	1.97^{a}
Increase by maximum (20%) quinoa flour replacement (%)	27.10	68.01	21.20	57.30	74.00	11.20

**: $p< 0.01$; *: $p < 0.05$; ns: not significant; The means with the same letter in column are not significantly different ($p <0.05$)

raised mineral content of the bread samples. Bread samples containing 20% quinoa flour had the highest values of Ca, K, Fe, Mg, Mn and Zn minerals.

Stikic *et al.* (2012) determined Ca, K, Mg, Fe, Mn and Zn values as 60, 288, 45,1.77, 0.81 and 1.19 mg/100g, respectively in 20% quinoa flour replaced breads. These were followed by 15% and 10% quinoa flour addition. The breads prepared with 0% quinoa flour (control group) contained the lowest levels of these minerals. This was an expected result. It was reported by many studies that quinoa seed and flour is a very rich source of major and minor minerals. Thus, the use of quinoa flour instead of wheat flour, which has a high content of minerals, led to an increase in mineral content of the final product.

***Conclusion*:** According to the results, phytic acid, TPC and mineral content of quinoa flour were found higher than wheat flour. In recent years much attention has been given to natural phenolics which plays an important role in scavenging oxidative radicals. Especially, Tosunbey wheat variety was found to have positive effects on phytic acid content. As conclusion, the use of quinoa flour in the production of bread was found suitable for the improvement of nutritional properties. To consider nutritional properties of bread, the best combination was found as Tosunbey and 20% quinoa flour in terms of mineral, phytic acid and TPC content. In addition to being superior to nutritional value, it is a drought-resistant and adaptable plant to many climatic conditions, which can be produced as an alternative to plants that are more water-demanding like rice and can be added to various food formulations.

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