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Gluten-free tarhana fortified with different ratios of edible mushroom *Lactarius deliciosus*

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Abstract

The present work aimed to develop nutritious and delicious tarhana soup for people who are especially sensitive to gluten. An edible mushroom, *Lactarius deliciosus*, was used in the gluten-free soup formulation instead of rice flour with different fortification ratios (0, 25, 50, 75, and 100%). Crude ash, crude protein, crude fat, mineral, acidity, water and oil absorptions, total phenolics, and antioxidant activities of gluten-free tarhana samples increased with the fortification of *L. deliciosus* in a dose-dependent manner, though the samples lost their lightness (*L**), hue angle (H°), and chroma (C*) values. Total carbohydrate was calculated by subtracting the sum of crude ash, crude protein, and crude fat contents, and and determined in the range of 64.31 - 78.51%. Potassium was the most abundant mineral found in samples, followed by calcium, and magnesium. Total polyphenols increased to 14,847.28 from 1,526.46 mg/kg gallic acid equivalent (GAE) for 100% fortification of *L. deliciosus* powder on dry weight basis (DWB). Antioxidant activity by ferric reducing antioxidant power (FRAP) assay (3.01 - 10.14 mmol Trolox equivalent, TE/kg DWB) was comparably higher than that with the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay (1.09 - 8.81 mmol TE/kg DWB). Gluten-free tarhana sample fortified with 25% *L. deliciosus* powder (DWB) had the highest sensory scores with respect to colour, taste, mouthfeel, and overall acceptability.

Introduction

Tarhana is a traditional fermented Turkish product, generally consists of wheat flour, yoghurt, tomato and/or pepper paste, yeast (*Saccharomyces cerevisiae*), and spices. Home-made tarhana is more common, but on an industrial scale, the production of ready-to-eat tarhana product has become more popular recently (Maskan and Ibanoğlu, 2002; Bilgicli, 2009a). Tarhana has a high nutritional value, for it contains essential amino acids such as valine, leucine, isoleucine, methionine, and phenylalanine. Wheat, however, is not suitable for people with celiac, baker's asthma, and gluten sensitivity. Gluten, which is a fundamental protein component of dough, creates an excellent viscoelasticity, thereby improving the nutritional and technological properties of the product. In gluten-free products, rice flour is mostly used due to its high starch content (Hager *et al.,* 2012).

Mushrooms have gained popularity in recent years as a type of functional food due to their proteins, vitamins, and mineral contents, as well as secondary metabolites that are useful for therapeutic purposes (Kosanić *et al.,* 2016). *Lactarius deliciosus*, commonly known saffron milk cap, belongs to family Russulaceae, and is a well-known wild edible mushroom, growing mostly under pine trees (Adanacioglu *et al.,* 2007). Its antioxidant activity has been reported as comparably higher as compared to other edible mushrooms (Orhan and Ustun, 2011; Palacios *et al.,* 2011). Unfortunately, edible mushrooms typically have a storage life of only three days at 25°C due to their high moisture levels and enzyme activities. Thermal operations such as drying and deep-freezing could prolong storage stabilities and help to preserve nutrient contents (Barros *et al.,* 2007). Nowadays, researchers are more focused on developing food products using certain parts, extracts, or powders of mushrooms in order to benefit from their nutritional advantages. *Agaricus bisporus* (portobello mushroom) and *Lentinula edodes* (shiitake mushroom) are the most used types in functional food formulations. Bread (Ahmad and Singh, 2016), sponge cake (Arora *et al.,* 2017), and noodle (Kim *et al.,* 2009) have been reported as novel mushroom-fortified food products.

The present work developed an alternative gluten-free tarhana formulation using *L. deliciosus* powder to improve its nutritional and functional

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qualities. The fortification of tarhana has been done with buckwheat flour (Bilgicli, 2009b), oat flour (Degirmencioglu *et al.,* 2016), barley (Erkan *et al.,* 2006), wheat germ/bran (Bilgicli *et al.,* 2006), quinoa flour (Demir, 2014), cherry laurel (Temiz and Tarakci, 2017), and carob flour (Herken and Aydin, 2015).

Materials and methods

Materials

Rice flour, full-fat cow milk yoghurt, tomato paste, pepper paste, onion, compressed baker's yeast (*S. cerevisiae*), and table salt (NaCl) were purchased from a local market in Osmaniye, Turkey. *L. deliciosus* (voucher no: FBozok00300) were collected from Akyar Village (37°01'08" N, 36°13'49" E, 150 m), Osmaniye, Turkey. All parts of mushrooms were dried using a dehydrator (KYS 329A; Guandong Kangye Electric Appliance Co., Ltd., China) at 40°C for 1 d, and powdered using a blender (Waring, Germany). All chemicals and reagents used were purchased from Sigma Aldrich (USA).

Gluten-free tarhana production

Rice flour (100 g), yoghurt (50 g, fat content = 3.8%), pepper paste (7.5 g), tomato paste (2.5 g), onion (2.5 g), salt (7.5 g), and yeast (2 g) were mixed using a mixer (KitchenAid, USA) for 5 min at the highest speed to prepare the control sample (modified from Kilci and Gocmen, 2014). For *L. deliciosus*-fortified samples, rice flour was substituted with 25, 50, 75, and 100% (w/w) *L. deliciosus* powder. The dough was allowed to incubate at room temperature (25°C) for 72 h, and dried until the moisture content reached below 10%. The dried mixture was milled using a blender (Waring, Germany), and stored at 4°C until further analysis.

Colour analysis

Colour was measured using a portable colorimeter (Konica Minolta CR 400, Japan) based on the CIELab colour space system (L^*: 0 = black, 100 = white; a^*: + red, - green; b^*: + yellow, - blue). Chroma (C*) and hue angle (H°) were calculated using Eqs. 1 and 2, respectively. Values are the mean of three determinations.

$$C^* = \sqrt{(a^*)^2 + (b^*)^2} \quad \text{(Eq. 1)}$$

$$H^\circ = \tan^{-1}\left(\frac{b^*}{a^*}\right) \quad \text{(Eq. 2)}$$

Chemical analysis

AACC methods for crude ash (method 08-01), crude protein (method 46012), and crude fat (method 30-25) were performed. The mineral content was determined using an atomic absorption spectrometry (Agilent, GTA 120, USA) (Podio *et al.,* 2013). The total carbohydrate were determined using Eq. 3 (Vaz *et al.,* 2011), and the results of related nutrients were given on dry weight basis (DWB) as mean value of three measurements:

Total carbohydrate (%) = 100% – (protein% + fat% + ash%) (Eq. 3)

The pH values were determined by a digital pH meter (Thermo Scientific, Orion star 3, USA) (5 g of gluten-free tarhana sample in 50 mL of distilled water), and the total titratable acidity was expressed as lactic acid following the procedure of Kirk and Sawyer (1991). Data were the mean value of three measurements.

Water and oil absorption capacity

Gluten-free tarhana sample (5 g) was mixed with 25 mL of distilled water or sunflower oil, and then the mixtures were stirred vigorously. Centrifugation of samples (Hettich Zentrifugen, Germany) was performed at 6,000 rpm for 10 min, and the mixture was filtered through Whatman No. 1 filter paper. The volume of supernatant was measured, and water and oil absorption capacities were stated as mL of water or mL of oil/g gluten-free tarhana (Bilgicli, 2009a). Data were the mean value of three measurements.

Preparation of extract and determination of total polyphenols and antioxidant activities

Gluten-free tarhana sample (1 g) was mixed with 20 mL of distilled water, and vortexed (Wisd Vortex Mixer VM-10; Germany) for 15 s. The mixture was then sonicated in an ultrasonic water bath (Selecta, Germany) at 25°C for 20 min, and then centrifuged (Hettich Zentrifugen, Germany) at 3,500 rpm for 15 min. The extracts were then filtered through Whatman No. 1 filter paper, and stored at 4°C (Bennett *et al.,* 2011). The method of Li *et al.* (2015) was used to determine the total polyphenols, and results were expressed as mg of gallic acid equivalent, GAE per kg DWB. The antioxidant activities were analysed using the DPPH and FRAP assays according to Aghraz *et al.* (2018) and Szydłowska-Czerniak *et al.* (2008), respectively. Results were expressed as mmol Trolox equivalent, TE per kg DWB. Data were the mean value of three measurements.

Sensory analysis

Seven trained panellists from the Department of Food Engineering, Faculty of Engineering, Osmaniye Korkut Ata University who were familiar with both tarhana and mushroom evaluated the soup samples. Briefly, gluten-free tarhana (20 g) was dissolved in 200 mL of pure water, and cooked with constant stirring. After 2 min of boiling, 2 g of butter was added, and the soup was cooled down to nearly 70°C, and served in a porcelain bowl (Kilci and Gocmen, 2014). Samples were randomly coded with 3-digit numbers. The panellists evaluated the colour, odour, taste, mouthfeel, and overall acceptability of the samples by using the nine-point hedonic scale (1 = dislike extremely, 5 = acceptable, and 9 = like extremely).

Statistical analysis

The experimental data were analysed by the SPPS software (trial version 18.0, USA) using an analysis of variance (ANOVA, one way) at 95% confidence interval. Duncan's test was used to determine homogenous groups. Pearson's correlation (at both 0.05 and 0.01 significance levels, 2-tailed) was performed to assess the linear relation between two random variables, and principal component analysis (PCA) was conducted on standardised data.

Results and discussion

Colour analysis

The colour values of gluten-free tarhana samples are shown in Table 1. The fortification of *L. deliciosus* powder into the soup formulation affected the lightness (L^*) of product significantly ($p < 0.05$). As the fortification ratio increased, the L^*, b^*, C*, and H° values of specimens decreased. Although fresh *L. deliciosus* did not have a dark colour, drying (in order to obtain powder form) caused both non-enzymatic (Das and Arora, 2018) and enzymatic browning reactions in mushrooms. High mineral content of *L. deliciosus* could also help to catalyse non-enzymatic browning (Bilgicli, 2009a). The strong and negative correlations between L^* and potassium (K), calcium (Ca), magnesium (Mg), copper (Cu), manganese (Mn), iron (Fe), and zinc (Zn) may support this assertion ($p < 0.01$). The other reason for this could be due to the degradation of polysaccharides (Bilgicli, 2009b), coming from rice flour and/or *L. deliciosus* powder. At 50% *L. deliciosus* fortification, the a^* value of gluten-free tarhana decreased, but at 75 and 100% *L. deliciosus* fortification, a^* increased, but not exceeding 8.20 (0%). Furthermore, there were negative relationships between L^* and both polyphenols (r = -0.96; $p < 0.01$) and antioxidant activities by DPPH and FRAP assay (r = -0.97 and r = -0.99; $p < 0.01$). Therefore, it could be assumed that, bioactive compounds contributed to the product's lightness.

Chemical analysis

Table 1 shows the chemical composition of gluten-free tarhana samples on dry weight basis (DWB). The crude ash, crude protein, crude fat, mineral contents, and acidity of gluten-free tarhana samples increased with increasing *L. deliciosus* powder in the formulation. This is most likely due to the high levels of related nutrients and acidity in mushrooms. Kalogeropoulos *et al.* (2013) determined the crude compositions of *L. deliciosus, L. sanguifluus, L. semisanguifluus, Russula delica*, and *Suillus bellinii* collected from Greece, and reported that the lowest moisture content was in *L. deliciosus*, which meaned that, it had the highest dry matter/nutritive value (11.05%), as well as carbohydrates (8.55%).

The ash level of control (6.67%) was in line with Daglioglu (2000) who reported that the average ash contents of several tarhana were nearly 6.20%. *L. deliciosus* has high ash content (Altuntas *et al.,* 2016), and gluten-free tarhana samples containing *L. deliciosus* powder had ash in the range of 7.87 - 11.05%. The crude protein level of gluten-free tarhana samples increased from 11.92 to 20.18% (100% *L. deliciosus* powder fortification). Therefore, the newly formulated gluten-free tarhana could be a good source of protein for human diet.

The crude fat level did not differ significantly ($p > 0.05$), and the fat level was less than other gluten-free tarhana reported by Bilgicli (2009b) and Demir (2014). The increasing ratio of *L. deliciosus* powder and augmentation of fat, ash, and protein decreased the carbohydrate level of gluten-free tarhana samples in the present work. Also, rice flour may contain more carbohydrates than *L. deliciosus* powder. Carbohydrates are the major nutrient group in mushroom species, and β-glucan and chitin comprise significant part of the cell wall polysaccharides. These compounds are classified as 'water insoluble dietary fibre', and cannot be digested by enzymes in humans. The biopharmacological effects of mushroom sclerotium, which has a higher content of β-glucan, include inducing apoptosis of leukemic cells, antitumor activity, and hepatic protection (Cheung, 2013). Furthermore, dietary fibres are known for their lower calories than other carbohydrates (Westenbrink *et al.,* 2013). Therefore, *L. deliciosus* could be a functional ingredient in

Table 1. Nutrients (on DWB), functional properties, colour values, and antioxidant activities of gluten-free tarhana samples fortified with *L. deliciosus* powder at different percentages.

	0%	25%	50%	75%	100%
Crude ash (%)	6.67 ± 0.09^{d}	7.87 ± 0.13^{c}	9.90 ± 0.27^{b}	10.93 ± 0.49^{a}	11.05 ± 0.04^{a}
Crude protein (%)	11.92 ± 0.58^{c}	15.08 ± 0.84^{b}	19.17 ± 0.85^{a}	19.84 ± 0.05^{a}	20.18 ± 0.15^{a}
Crude fat (%)	2.90 ± 0.01^{a}	3.08 ± 0.76^{a}	4.09 ± 0.43^{a}	4.24 ± 1.26^{a}	4.45 ± 1.11^{a}
Total carbohydrate (%)	78.51 ± 0.49^{a}	73.97 ± 1.46^{b}	66.84 ± 1.01^{c}	64.98 ± 1.75^{c}	64.31 ± 1.00^{c}
K (mg/kg)	2,732.69 ± 45.33^{e}	6,294.47 ± 186.00^{d}	9,376.89 ± 121.41^{c}	15,167.94 ± 500.58^{b}	16,497.08 ± 636.03^{a}
Ca (mg/kg)	759.45 ± 0.12^{c}	796.64 ± 0.02^{bc}	851.74 ± 95.33^{ab}	867.59 ± 0.05^{ab}	898.75 ± 6.93^{a}
Mg (mg/kg)	208.21 ± 25.32^{e}	345.35 ± 33.33^{d}	400.33 ± 10.65^{c}	587.87 ± 18.77^{b}	674.43 ± 11.31^{a}
Cu (mg/kg)	0.96 ± 0.10^{c}	3.39 ± 1.88^{b}	3.43 ± 0.94^{b}	4.86 ± 0.63^{ab}	5.84 ± 0.82^{a}
Mn (mg/kg)	8.74 ± 1.05^{e}	20.69 ± 0.61^{d}	29.64 ± 0.51^{c}	45.81 ± 3.27^{b}	59.00 ± 0.82^{a}
Fe (mg/kg)	10.16 ± 1.35^{e}	96.72 ± 8.47^{d}	158.94 ± 10.29^{c}	289.75 ± 13.15^{b}	422.04 ± 5.66^{a}
Zn (mg/kg)	16.56 ± 0.22^{e}	33.42 ± 0.44^{d}	41.82 ± 0.58^{c}	63.52 ± 0.47^{b}	76.88 ± 1.31^{a}
pH	4.69 ± 0.01^{d}	5.09 ± 0.01^{c}	5.23 ± 0.01^{b}	5.29 ± 0.02^{a}	5.22 ± 0.01^{b}
Acidity (%)	0.60 ± 0.02^{e}	1.13 ± 0.01^{d}	1.41 ± 0.01^{c}	1.74 ± 0.01^{b}	1.97 ± 0.01^{a}
Water absorption (ml/g)	0.53 ± 0.04^{b}	0.68 ± 0.08^{b}	0.84 ± 0.25^{b}	1.24 ± 0.06^{a}	1.59 ± 0.13^{a}
Oil absorption (ml/g)	0.99 ± 0.27^{a}	1.19 ± 0.21^{a}	1.20 ± 0.11^{a}	1.30 ± 0.42^{a}	1.30 ± 0.42^{a}
*L**	82.04 ± 0.93^{a}	64.22 ± 1.16^{b}	51.97 ± 0.39^{c}	36.62 ± 0.58^{d}	30.47 ± 0.46^{e}
*a**	8.20 ± 0.41^{a}	5.48 ± 0.46^{bc}	5.23 ± 0.20^{c}	5.88 ± 0.21^{bc}	6.13 ± 0.77^{b}
*b**	38.63 ± 0.58^{a}	25.65 ± 2.13^{b}	19.84 ± 0.39^{c}	18.45 ± 0.59^{cd}	17.35 ± 1.25^{d}
C*	39.49 ± 0.64^{a}	26.23 ± 2.17^{b}	20.51 ± 0.41^{c}	19.36 ± 0.62^{c}	18.40 ± 1.41^{c}
H°	78.02 ± 0.46^{a}	77.94 ± 0.25^{a}	75.24 ± 0.36^{b}	72.31 ± 0.10^{c}	70.57 ± 1.16^{d}
Total polyphenols (mg GAE/kg DW)	1,526.46 ± 66.76^{e}	4,761.38 ± 179.74^{d}	5,802.68 ± 63.07^{c}	13,838.10 ± 56.48^{b}	14,847.28 ± 00.10^{a}
DPPH (mmol TE/kg DW)	1.09 ± 0.10^{e}	5.70 ± 0.07^{d}	6.72 ± 0.05^{c}	8.54 ± 0.12^{b}	8.81 ± 0.11^{a}
FRAP (mmol TE/kg DW)	3.01 ± 1.11^{c}	5.78 ± 0.65^{b}	6.95 ± 0.05^{b}	9.77 ± 0.47^{a}	10.14 ± 0.93^{a}

Values are mean ± standard deviation of triplicate (n = 3). Means in a row with different lowercase superscripts are significantly different according to one way ANOVA (Duncan) test (p > 0.05). DPPH: 2,2-diphenyl-1-picrylhydrazyl; FRAP: ferric reducing antioxidant power; GAE: gallic acid equivalent; TE: Trolox equivalent; and DWB: dry weight basis.

gluten-free tarhana useful for weight regulation / management.

K, Ca, and Mg are the major minerals, while Cu, Mn, Fe, and Zn are the minor minerals that play important roles in human body such as strengthening bones and teeth, transmitting nerve impulses, and involved in hormones (Gharibzahedi and Jafari, 2017). The fortification of *L. deliciosus* powder significantly enhanced the mineral levels ($p < 0.05$). K was the highest mineral which varied between 2,732.69 and 16,497.08 mg/kg in gluten-free tarhana samples. Tarhana has been reported as a good source of minerals by Degirmencioglu *et al.* (2016), and our results agree with this claim. Moreover, Ca showed the strongest relationship with ash, protein, fat, and carbohydrate at 1% significance level in comparison with other micronutrients. Ca could comprise the biggest part of ash and bind proteins and fatty acids. Besides, *L. deliciosus* powder had 9.51- and 42.20-fold additive effects on Fe amounts of control and 100% fortification of *L. deliciosus*, respectively. Kosanić *et al.* (2016) reported that, Fe was the superior minor mineral found in *L. deliciosus*, and their findings are compatible with the present work.

The pH values which are important for sensorial characteristics were recorded as 4.69, 5.09, 5.23, 5.29, and 5.22 for gluten-free tarhana samples fortified with 0, 25, 50, 75, and 100% of *L. deliciosus* powder, respectively. Similarly, higher fortification with *L. deliciosus* powder resulted in higher acidity levels. There was also a close relationship between pH and acidity with respect to Pearson's correlation analysis ($r = 0.90$; $p < 0.05$). Generally, there is an inverse proportion between pH and acidity; but, in our study, this assertion is mostly invalid. The possible reason is that, titratable acidic substances may partially dissociate in aqueous medium during measurements. On the other hand, the buffering effect of mushroom proteins caused no important changes in pH (Bilgicli *et al.,* 2006), especially in samples with high fortification ratios (≥ 50%).

Water and oil absorption capacity

Water absorption plays an important role in viscous foods, and oil absorption acts by preventing phase separation as well as hydrophobicity degree (Ertas *et al.,* 2015). Table 1 shows the functional features of gluten-free tarhana samples and that 100% fortification of *L. deliciosus* powder increased water and oil absorption of gluten-free tarhana solids from 0.53 to 1.59, and 0.99 to 1.30 mL/g, respectively. No statistical significance was observed in oil absorptions ($p > 0.05$), and all gluten-free tarhana samples (except 100%) absorbed more oil than water. Increased water absorption can be attributed to the increased tendency of protein as *L. deliciosus* ratio was increased. Some studies reported water absorptions (on average) lower than our findings, as reported by Bilgicli (2009a) for tarhana with buckwheat flour (between 0.50 - 0.63 mL/g) and by Ertas *et al.* (2015) for tarhana enriched with whey concentrate (between 0.41 - 0.73 mL/g). El-Adawy and Taha (2001) also reported that, high water and oil absorption is advantageous in some flour products as thickening agents in soups or meat replacers. For this reason, *L. deliciosus* powder may be accepted as a useful choice.

Total polyphenols and antioxidant activities

The levels of total polyphenols and antioxidant activities of gluten-free tarhana samples are presented in Table 1. When compared with the control sample, total polyphenols and antioxidant activities (from both assays) of all gluten-free tarhana samples increased gradually with increasing fortification of *L. deliciosus*. The amounts of total polyphenols significantly increased ($p < 0.05$) in the range of nearly 32 - 102% with fortifications of *L. deliciosus* powder. Previous antioxidant studies in *L. deliciosus* are in close agreement with the present work. Fernandes *et al.* (2013) reported that the phenolic content of Portuguese freeze-dried *L. deliciosus* (in methanolic extract) was 24,000 mg GAE/kg. Furthermore, Ferreira *et al.* (2007) stated that the entire mushroom had higher polyphenols than the stipe or the cap. In the present work, all parts of *L. deliciosus* were dried and used in producing the gluten-free tarhana. The remarkably positive correlations ($p \leq 0.01$) between polyphenols and Mg (**r** = 0.99), Fe ($r = 0.97$), Zn ($r = 0.98$), and Mn ($r = 0.98$) were observed (Figure 1A). The determination coefficients (R^2), which expressed the relationship between polyphenols and Mg, Fe, Zn, and Mn were 0.98, 0.94, 0.97, and 0.95, respectively. It has also been stated that, phenolics had a significant impact on the development of metal ion complexes (Plizska, 2020), and minerals might probably be part of bioactive substances. Sulaiman *et al.* (2011) indicated that, minerals could have a role in activating enzymes which are used for biosynthesis of polyphenols. Moreover, Zn and Mn can be co-factors in an antioxidant enzyme - superoxide dismutase, which transforms superoxides to hydrogen peroxides (Fukai and Ushio-Fukai, 2011).

A 25% fortification of *L. deliciosus* powder increased the antioxidant activities by 5.23- and 1.92-fold in the DPPH and FRAP assays, respectively, when compared with control. Such

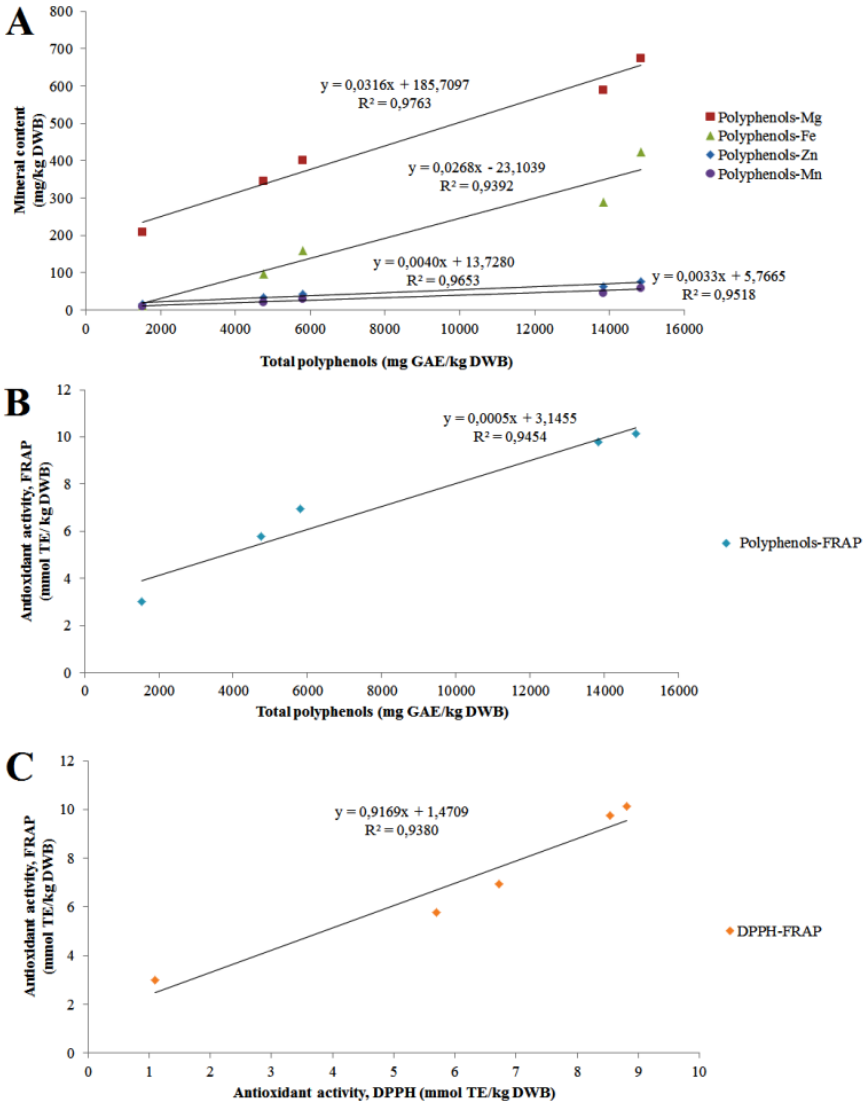

Figure 1. Linear correlations between some selected test results of tarhana: (A) phenolics-Mg (r = 0.99, R^2 = 0.98), phenolics-Fe (r = 0.97, R^2 = 0.94), phenolics-Zn (r = 0.98, R^2 = 0.97), phenolics-Mn (r = 0.98, R^2 = 0.95); (B) phenolics-antioxidant activity, FRAP (r = 0.97, R^2 = 0.95); and (C) antioxidant activity, DPPH-antioxidant activity, FRAP (r = 0.97, R^2 = 0.94) at 99% confidence interval.

rates tended to increase progressively in higher *L. deliciosus* powder concentrations. Jayakumar *et al.* (2009) attributed the relation between increased concentration and reducing power (Fe^{+3} - Fe^{+2} transformation) to the availability of reductones in medicinal mushrooms. Reductones (*e.g.* ascorbic acid) can deactivate free radicals and prevent peroxides. Figures 1B and 1C illustrate the relevance between polyphenols-FRAP and DPPH-FRAP. Figure 1B explains that bioactive molecules which have the ability to reduce Fe^{+3} ions are also categorised in polyphenols group. Figure 1C demonstrates that, most of the antioxidants in gluten-free tarhana samples can be detected through either method.

Sensory analysis

Sensory perceptions of soups made from gluten-free tarhana with *L. deliciosus* fortification are exhibited in Figure 2. Although the impact of different ratios of *L. deliciosus* powder fortification on colour, odour, taste, mouthfeel, and overall acceptability of gluten-free tarhana was insignificant (p > 0.05), the use of *L. deliciosus* powder in gluten-free tarhana formulation had acceptable results when, the control sample was taken into account. Gluten-free tarhana soup fortified with 25% of *L. deliciosus* powder yielded the highest points in all sensorial attributes, except odour. Overall acceptability of gluten-free tarhana samples were in the following order: 25% > 0% > 75% > 50% >

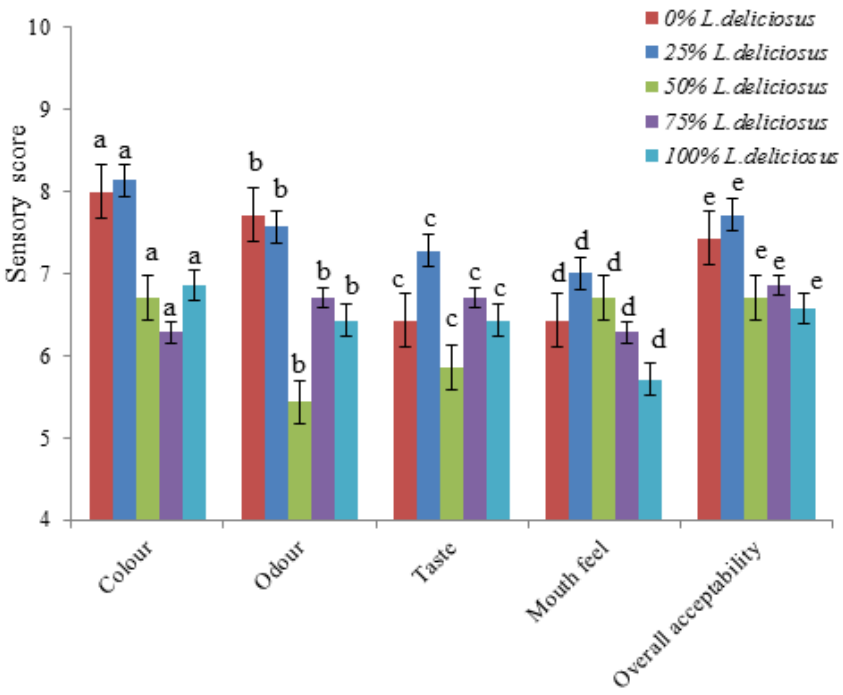

Figure 2. Sensory results of gluten-free tarhana samples fortified with *L. deliciosus* powder at different percentages. Similar lowercase in each group/parameter indicates no significant differences between samples according to one way ANOVA (Duncan) test ($p > 0.05$).

100%. An accurate trend of increasing or decreasing continuously with changing fortification ratios which was reported by Herken and Aydin (2015) and Ertas *et al.* (2009) was not observed in consumers' responses in the present work. Hence, *L. deliciosus* fortification did not have an undesirable taste in gluten-free tarhana soup. This is also consistent with the study of Kilci and Gocmen (2014) in tarhana supplemented with oat flour.

Principal component analysis

PCA was conducted on gluten-free tarhana samples to see whether the samples were grouped differently due to *L. deliciosus* fortification ratios, and the projections are given in Figure 3. The score plot of principal components accounts for 91.46% of dataset variance in two components: PC1 (81.69%) and PC2 (9.77%). According to the loading plot (Figure 3a) and distribution of scores (Figure 3b), specimens could be classified into three different

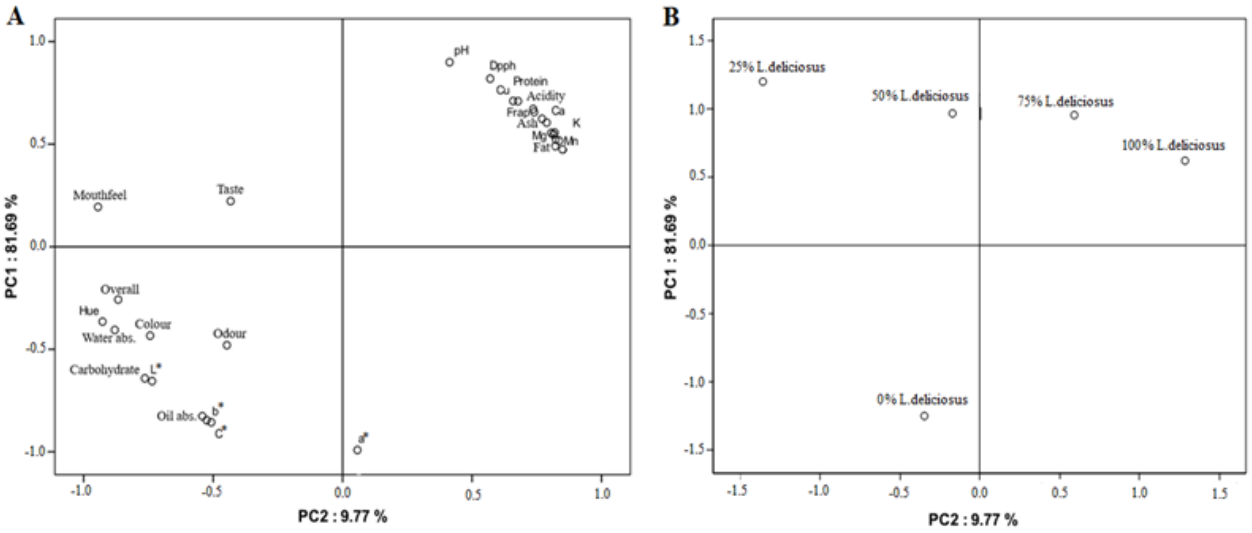

Figure 3. Score plot of principal component analysis (PCA) (A) illustrating the variation among the analysis, and (B) loading plot of PCA illustrating the variation among the gluten-free tarhana samples.

blocks: first block included both 25 and 50% *L. deliciosus*-fortified tarhana, second block included 75 and 100% *L. deliciosus*-fortified tarhana, and final block included the control sample. A clear division was identified between control and samples fortified with *L. deliciosus* powder, and except for the control, all samples were located on the positive axis of PC1 component. First block showed a lower abundance of mineral contents, pH, acidity, ash, fat, and antioxidant (DPPH and FRAP), however gluten-free tarhana fortified with 75 and 100% *L. deliciosus* powder were associated with a greater abundance of the aforementioned compounds. On the other hand, oil absorption, total carbohydrate, *L**, *b**, and C* were strongly correlated with the control sample as located on the negative axis of PC2. Mineral contents and the results of spectrophotometrical analysis depicted a negative relationship with colour parameters (except *a**), some of sensorial properties, and water/oil absorptions. Likewise, there was a negative relationship between *a** and two of sensorial attributes (mouthfeel and taste).

Conclusion

The purpose of the present work was to develop a gluten-free tarhana with an edible mushroom, *L. deliciosus*, for celiac patients and individuals who would like to consume alternative and healthier food. The fortification of the tarhana formulation with *L. deliciosus* powder instead of rice flour resulted in higher proteins (from 3.16 to 8.26%), minerals, phenolics (from 3.23 to 13.32 mg GAE/g DWB), and antioxidant activities (from 4.61 to 7.72 mmol TE/kg DWB for DPPH, and from 2.77 to 7.13 mmol TE/kg DWB for FRAP) as compared to the control sample. The most abundant macro- and microminerals were K (2,732.69 – 16,497.08 mg/kg) and Fe (10.16 - 422.04 mg/kg), respectively. Strong and significant correlations were determined among mineral contents, total polyphenols, and antioxidant activities ($p < 0.01$), with the R^2 values ranging from 0.94 to 0.97. Although appreciable differences were observed in the colour of samples, the panellists were not seen prejudiced. Based on the sensory results, fortification ratio of 25% could be recommended. Future research will be focused on the digestion and rheological behaviour of soup as well as investigating the dietary fibres that come from mushrooms like β-glucan and chitin which have the potential of offering a low-calorie diet.

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References

Adanacioglu, N., Tan, A., Karabak, S., Guzelsoy, N., Ayas, F., Aykas, L. and Taylan, T. 2007. Economically important wild mushroom saffron milk cap (*Lactarius deliciosus* (L.) Gray) of Aegean region, Turkey. Anadolu Journal of AARI 27(2): 91-96.

Aghraz, A., Gonçalves, S., Rodríguez-Solana, R., Dra, L. A., Di Stefano, V., Dugo, G., … and Romano, A., 2018. Antioxidant activity and enzymes inhibitory properties of several extracts from two Moroccan Asteraceae species. South African Journal of Botany 118: 58-64.

Ahmad, K. and Singh, N. 2016. Evaluation of nutritional quality of developed functional bread fortified with mushroom and dates. The Clarion 5(1): 23-28.

Altuntas, D., Alli, H., Kaplaner, E. and Ozturk, M. 2016. Determination of fatty acid constituents and macro-nutritional properties of some *Lactarius* species. Turkish Journal of Agriculture - Food Science and Technology 4(3): 216-220.

American Association of Cereal Chemists (AACC). 1990. Approved methods of the American Association of Cereal Chemists. 8th ed. United States: AACC.

Arora, B., Kamal, S. and Sharma, V. 2017. Sensory, nutritional and quality attributes of sponge cake supplemented with mushroom (*Agaricus bisporus*) powder. Nutrition and Food Science 47: 578-590.

Barros, L., Baptista, P., Correia, D. M., Morais, J. S. and Ferreira, I. C. F. R. 2007. Effects of conservation treatment and cooking on the chemical composition and antioxidant activity of Portuguese wild edible mushrooms. Journal of Agricultural and Food Chemistry 55(12): 4781-4788.

Bennett, L. E., Jegasothy, H., Konczak, I., Frank, D., Sudharmarajan, S. and Clingeleffer, P. R. 2011. Total polyphenolics and anti-oxidant properties of selected dried fruits and relationships to drying conditions. Journal of Functional Foods 3(2): 115-124.

Bilgicli, N. 2009a. Enrichment of gluten-free tarhana with buckwheat flour. International Journal of Food Science and Nutrition 60(S4): 1-8.

Bilgicli, N. 2009b. Effect of buckwheat flour on chemical and functional properties of tarhana.

LWT - Food Science and Technology 42(2): 514-518.

Bilgicli, N., Elgun, A., Herken, E. N., Selman, T., Ertas, N. and Ibanoglu, S. 2006. Effect of wheat germ/bran addition on the chemical, nutritional and sensory quality of tarhana, a fermented wheat flour-yoghurt product. Journal of Food Engineering 77(3): 680-686.

Cheung, P. C. K. 2013. Mini-review on edible mushrooms as source of dietary fiber: preparation and health benefits. Food Science and Human Wellness 2(3-4): 162-166.

Daglioglu, O. 2000. Tarhana as a traditional Turkish fermented cereal food. Its recipe, production and composition. Nahrung 44: 85-88.

Das, I. and Arora, A. 2018. Alternate microwave and convective hot air application for rapid mushroom drying. Journal of Food Engineering 223: 208-219.

Degirmencioglu, N., Gurbuz, O., Herken, E. N. and Yildiz, A. Y. 2016. The impact of drying techniques on phenolic compound, total phenolic content and antioxidant capacity of oat flour tarhana. Food Chemistry 194: 587-594.

Demir, M. K. 2014. Use of quinoa flour in the production of gluten-free tarhana. Food Science and Technology Research 20(5): 1087-1092.

El-Adawy, T. A. and Taha, K. M. 2001. Characteristics and composition of different seed oils and flours. Food Chemistry 74(1): 47-54.

Erkan, H., Celik, S., Bilgi, B. and Koksel, H. 2006. A new approach for the utilization of barley in food products: barley tarhana. Food Chemistry 97(1): 12-18.

Ertas, N., Sert, D. and Demir, M. K. 2015. Functional properties of tarhana enriched with whey concentrate. Agronomy Research 13(4): 919-928.

Ertas, N., Sert, D., Demir, M. K. and Elgun, A. 2009. Effect of whey concentrate on chemical, nutritional and sensory properties of tarhana. Food Science and Technology 15(1): 51-58.

Fernandes, Â., Antonio, A. L., Barreira, J. C. M., Botelho, M. L., Oliveira, M. B. P. P., Martins, A. and Ferreira, I. C. F. R. 2013. Effects of gamma irradiation on the chemical composition and antioxidant activity of *Lactarius deliciosus* L. wild edible mushroom. Food and Bioprocess Technology 6(10): 2895-2903.

Ferreira, I. C. F. R., Baptista, P., Vilas-Boas, M. and Barros, L. 2007. Free-radical scavenging capacity and reducing power of wild edible mushrooms from northeast Portugal: individual cap and stipe activity. Food Chemistry 100(4): 1511-1516.

Fukai, T. and Ushio-Fukai, M. 2011. Superoxide dismutases: role in redox signaling, vascular function, and diseases. Antioxidants and Redox Signaling 15: 1583-1606.

Gharibzahedi, S. M. T. and Jafari, S. M. 2017. The importance of minerals in human nutrition: bioavailability, food fortification, processing effects and nanoencapsulation. Trends in Food Science and Technology 62: 119-132.

Hager, A. S., Wolter, A., Jacob, F., Zannini, E. and Arendt, E. K. 2012. Nutritional properties and ultra-structure of commercial gluten free flours from different botanical sources compared to wheat flours. Journal of Cereal Science 56(2): 239-247.

Herken, E. N. and Aydin, N. 2015. Use of carob flour in the production of tarhana. Polish Journal of Food and Nutritional Science 65(3): 167-174.

Jayakumar, T., Thomas, P. A. and Geraldine, P. 2009. *In-vitro* antioxidant activities of an ethanolic extract of the oyster mushroom, *Pleurotus ostreatus*. Innovative Food Science and Emerging Technologies 10: 228-234.

Kalogeropoulos, N., Yanni, A. E., Koutrotsios, G. and Aloupi, M. 2013. Bioactive microconstituents and antioxidant properties of wild edible mushrooms from the island of Lesvos, Greece. Food and Chemical Toxicology 55: 378-385.

Kilci, A. and Gocmen, D. 2014. Phenolic acid composition, antioxidant activity and phenolic content of tarhana supplemented with oat flour. Food Chemistry 151: 547-553.

Kim, S. Y., Chung, S. I., Nam, S. H. and Kang, M. Y. 2009. Cholesterol lowering action and antioxidant status improving efficacy of noodles made from unmarketable oak mushroom (*Lentinus edodes*) in high cholesterol fed rats. Journal of the Korean Society for Applied Biological Chemistry 52: 207-212.

Kirk, R. S. and Sawyer, R. 1991. Pearson's composition and analysis of foods. England: Longman Scientific and Technical.

Kosanić, M., Ranković, B., Rančić, A. and Stanojković, T. 2016. Evaluation of metal concentration and antioxidant, antimicrobial, and anticancer potentials of two edible mushrooms *Lactarius deliciosus* and *Macrolepiota procera*. Journal of Food and Drug Analysis 24(3): 477-484.

Li, X., Wasila, H., Liu, L., Yuan, T., Gao, Z., Zhao, B. and Ahmad, I. 2015. Physicochemical characteristics, polyphenol compositions and antioxidant potential of pomegranate juices from 10 Chinese cultivars and the environmental

factors analysis. Food Chemistry 175: 575-584.
Maskan, M. and Ibanoğlu, S. 2002. Hot air drying of cooked and uncooked tarhana dough, a wheat flour–yoghurt mixture. European Food Research and Technology 215: 413–418.
Orhan, I. and Ustun, O. 2011. Determination of total phenol content, antioxidant activity and acetylcholinesterase inhibition in selected mushrooms from Turkey. Journal of Food Composition and Analysis 24(3): 386-390.
Palacios, I., Lozano, M., Moro, C., D'Arrigo, M., Rostagno, M. A., Martínez, J. A., ... and Villares, A. 2011. Antioxidant properties of phenolic compounds occurring in edible mushrooms. Food Chemistry 128(3): 674-678.
Plizska, 2020. Content and correlation of polyphenolic compounds, bioelements and antiradical activity in black elder berries (*Sambucus nigra* L.). Journal of Elementology 25(2): 595-605.
Podio, N. S., Baroni, M. V., Badini, R. G., Inga, M., Ostera, H. A., Cagnoni, M., ... and Wunderlin, D. A. 2013. Elemental and isotopic fingerprint of Argentinean wheat. matching soil, water, and crop composition to differentiate provenance. Journal of Agricultural and Food Chemistry 61: 3763-3773.
Sulaiman, S. F., Yusoff, N. A. M., Eldeen, I. M., Seow, E. M., Sajak, A. A. B. and Supriatno, O. K. L. 2011. Correlation between total phenolic and mineral contents with antioxidant activity of eight Malaysian bananas (*Musa* sp.). Journal of Food Composition and Analysis 24(1): 1-10.
Szydłowska-Czerniak, A., Dianoczkib, C., Recseg, K., 2008. Determination of antioxidant capacities of vegetable oils by ferric-ion spectrophotometric methods. Talanta 76: 899-905.
Temiz, H. and Tarakci, Z. 2017. Composition of volatile aromatic compounds and minerals of tarhana enriched with cherry laurel (*Laurocerasus officinalis*). Journal of Food Science and Technology 54(3): 735-742.
Vaz, J. A., Barros, L., Martins, A., Santos-Buelga, C., Vasconcelos, M. H. and Ferreira, I. C. F. R. 2011. Chemical composition of wild edible mushrooms and antioxidant properties of their water soluble polysaccharidic and ethanolic fractions. Food Chemistry 126(2): 610-616.
Westenbrink, S., Brunt, K. and Van Der Kamp, J. W. 2013. Dietary fibre: challenges in production and use of food composition data. Food Chemistry 140(3): 562-567.