

PREPARATION OF LACTIC ACID BACTERIA FERMENTED WHEAT-YOGHURT MIXTURES

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ABSTRACT

Background. Tarhana, a wheat-yoghurt fermented mixture, is considered as a good source of saccharides, proteins, some vitamins and minerals. Moreover, their preparation is inexpensive and lactic acid fermentation offers benefits like product preservation, enhancement of nutritive value and sensory properties improvement. The aim of this work was to evaluate changes of some chemical parameters during fermentation of tarhana, when the level of salt and amount of yoghurt used were varied. Some functional and sensory characteristics of the final product were also determined.

Material and methods. Chemical analysis included determination of pH, titrable acidity, content of reducing saccharides, lactic, acetic and citric acid. Measured functional properties of tarhana powder were foaming capacity, foam stability, water absorption capacity, oil absorption capacity and emulsifying activity. Tarhana soups samples were evaluated for their sensory characteristics (colour, odor, taste, consistency and overall acceptability).

Results. Fermentation of tarhana by lactic acid bacteria and yeasts led to decrease in pH, content of reducing saccharides and citric acid, while titrable acidity and concentration of lactic and acetic acid increased. Determination of functional properties of tarhana powder showed, that salt absence and increased amount of yoghurt in tarhana recipe reduced foaming capacity and oil absorption capacity, whereas foam stability and water absorption capacity were improved. Sensory evaluation of tarhana soups showed that variations in tarhana recipe adversely affected sensory parameters of final products.

Conclusion. Variations in tarhana recipe (salt absence, increased proportion of yoghurt) led to changes in some chemical parameters (pH, titrable acidity, reducing saccharides, content of lactic, acetic and citric acid). Functional properties were also affected with changed tarhana recipe. Sensory characteristics determination showed, that standard tarhana fermented for 144 h had the highest overall acceptability.

Key words: tarhana, organic acids, functional properties, sensory characteristics

INTRODUCTION

Tarhana is a traditional Turkish fermented food product. It is produced by mixing wheat flour, yoghurt, yeast, vegetables (tomatoes, onions, green and red peppers), salt and spices (including mint and thyme), followed by fermentation for 1-7 days at room temperatures around 25-30°C [Gocmen et al. 2004, Sengun

et al. 2009]. Tarhana fermentation is usually carried out by using yoghurt bacteria, *Lactobacillus bulgaricus*, *Streptococcus thermophilus*, and bakers' yeast (*Saccharomyces cerevisiae*) [Lar et al. 2012].

The dough at fermentation is called wet tarhana. Afterwards, the dough is dried in sun or by dryer

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as a lump, nugget or thin layers to obtain dry tarhana. Finally, it is ground to powder < 1 mm. Tarhana is usually reconstituted with water and served as a hot soup. Tarhana soup can be prepared from wet or dry tarhana [Erbaş et al. 2005, Koca et al. 2002].

Tarhana is a very nutritive food because of nutritional deficiency in wheat mostly eliminated by yoghurt. Its nutritional value is increased and digestion is facilitated by fermentation [Dalgıç and Belibağh 2008]. The protein, carbohydrate and lipid components of tarhana mix are subjected to partial digestion and hydrolysis by lactic acid bacteria and yeasts during fermentation, resulting in a product with improved digestive properties [Tamer et al. 2007]. Tarhana is a good source of calcium, iron and zinc, as well as some other minerals [Daglioğlu 2000]. Fermentation results in significant increases of riboflavin, niacin, panthothenic acid, ascorbic acid and folic acid contents of tarhana [Bilgiçli 2009].

The low moisture content (3-9%) and low pH value (4.0-4.5) of the final product provide bacteriostatic effect against pathogenic and spoilage microorganisms and increase the shelf life of the product [Sengun and Karapinar 2012].

There are similar products with different names such as kishk in Egypt, Syria, Lebanon and Jordan, kushuk in Iraq, trahana in Greece and tahonya/talkuna in Hungary and Finland [Maskan and Ibanoglu 2002].

The purpose of this work was to evaluate the changes in some chemical parameters during fermentation process of tarhana samples. Tested samples had changed the amount of added salt and yoghurt compared to the control, due to both, salt and yoghurt may considerably affect microbial activity (a_w , population of lactic acid bacteria in yoghurt) but also sensory and functional properties. Sensory analysis of tarhana soups and functional properties of tarhana powder were also determined.

MATERIAL AND METHODS

Commercial wheat flour type T650 (Penam Nitra, Slovakia) with the moisture content of 13.16%, protein content of 11.07%, and ash content of 0.43% on dry basis was used. The white yoghurt (Rajo Bratislava, Slovakia) used was full fat 3.50% made from bovine milk. Onion, tomato puree, salt, baker's yeast

Table 1. Composition of tarhana recipes

Ingredient	Weight, g		
	T1	T2	T3
Wheat flour	1 000	1 000	1 000
Yoghurt	500	500	1000
Tomato puree	120	120	120
Onion	120	120	120
Salt	40	0	40
Baker's yeast	20	20	20
Sweet paprika	20	20	20
Dill	2	2	2
Mint	2	2	2

in active dry form, sweet paprika powder, dill powder and mint powder were purchased from local markets in Bratislava, Slovakia. The ingredients used in tarhana preparation are listed in Table 1.

Preparation of tarhana. Tarhana samples (control – T1, without added salt – T2 and with increased amount of yoghurt – T3) were prepared according to the method described by Erkan et al. [2006] with some modifications. To prepare tarhana samples, onions were chopped, sliced to small pieces and blended for 30 s with 50 cm³ of tap water. The tomato puree, salt, sweet paprika powder, dill powder and mint powder were added, blended for 30 s, brought to the boil and blended for 10 min. The mixture was left to cool to room temperature and then wheat flour, yoghurt and baker's yeast was added and the mixture was kneaded after the addition of a further 100 cm³ of tap water. The resulting mixture was taken into covered containers and incubated in a thermostat at 30 ±1°C for 144 h fermentation. During fermentation, samples were withdrawn at pre-specified intervals for analytical determinations.

Chemical analyses. The following analytical parameters were studied during fermentation: pH, titratable acidity, reducing saccharides according to Schoorl [Kohajdová and Karovičová 2005] and content of organic acids. Lactic, acetic and citric acid were determined by using the method of capillary isotachopheresis [Kohajdová et al. 2006]. Isotachophoretic

measurements were performed by using an isotachophoretic analyser ZKI 01 (Villa Labeco, Spišská Nová Ves, Slovak Republic), equipped with conductivity detector and two-line recorder TZ 4200 (Laboratorní přístroje, Prague, Czech Republic).

Functional properties. Tarhana powder was obtained by drying wet tarhana dough in air oven at 50°C for 48 hours and then ground and sieved to pass a 1 mm screen and was used for the determination of functional properties. The methods described by Hayta et al. [2002] were used for determination of foaming capacity (FC), foam stability (FS), water absorption capacity (WAC), oil absorption capacity (OAC) and emulsifying activity (EA) of tarhana powder samples.

Sensory evaluation of tarhana soups. Sensory evaluation of tarhana soups prepared from wet tarhana doughs fermented for 72 and 144 hours was performed by method Isik and Yapar [2012]. Tarhana soups were prepared by mixing 40 g tarhana sample with 500 cm³ water and simmering for 10 min with constant stirring. The cooked samples were served to the panelist at 80°C in random order [Erkan et al. 2006]. The 9 instructed assessors evaluated: colour, aroma, taste, consistency and overall acceptance on hedonic scale from 1 (dislike extremely) to 7 (like extremely).

Statistical analysis. All analyses were carried out using three independent determinations and expressed as mean \pm standard deviation. One-way analysis of variance (ANOVA) and Fisher's least significant

difference (LSD) multiple range test were applied to data (functional properties, sensory characteristics) to establish significance of differences between samples. Stagraphic Plus, Version 3.1 (Statsoft; Tulsa, Oklahoma, USA) was used as statistical analysis software.

RESULTS AND DISCUSSION

From the results it was concluded, that during fermentation the pH value (Fig. 1 A) of the tarhana dough samples decreased, whereas acidity (Fig. 1 B) increased. Production of acids by lactic acid bacteria of yoghurt and bakery yeast in the formulation during fermentation was the main reason of pH reduction in tarhana dough samples [Çelik et al. 2010].

Significant increase in acidity during the fermentation was observed in tarhana samples T2 (0.40-1.07%) and T3 (0.44-1.06%) compared to the control T1 (0.39-0.95%). According to Ibanoglu et al. [1999] if salt is added before fermentation, the fermentation activity and lactic acid concentration are lower when compared with the same composition of tarhana without salt. This could be explained by the higher water activity of the fermenting medium in which less salt is present to bind some of free water available. This could favour the growth of lactic acid bacteria [Ibanoglu et al. 1999]. Similar decrease in pH and increase in acidity as was presented in this study was also reported by Ibanoglu et al. [1995] and Ibanoglu et

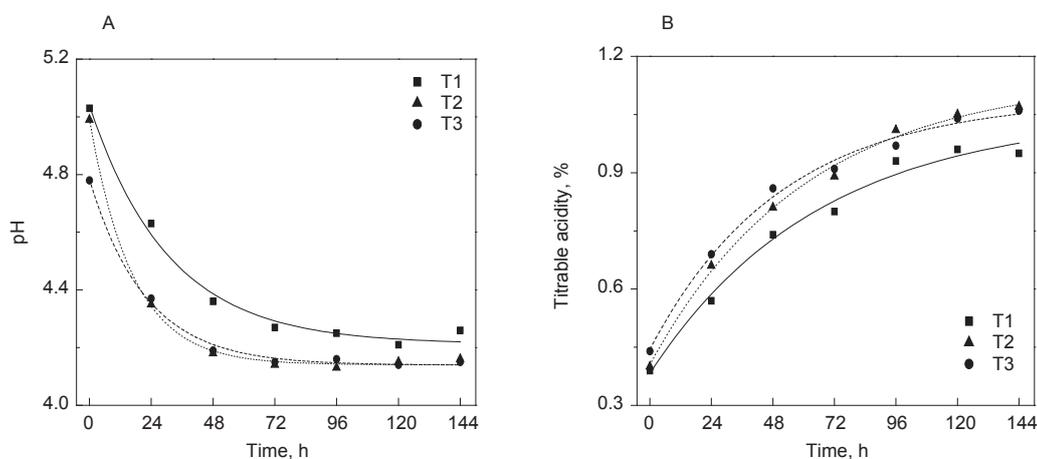


Fig. 1. Changes in pH (A) and titrable acidity (B) during tarhana samples fermentation

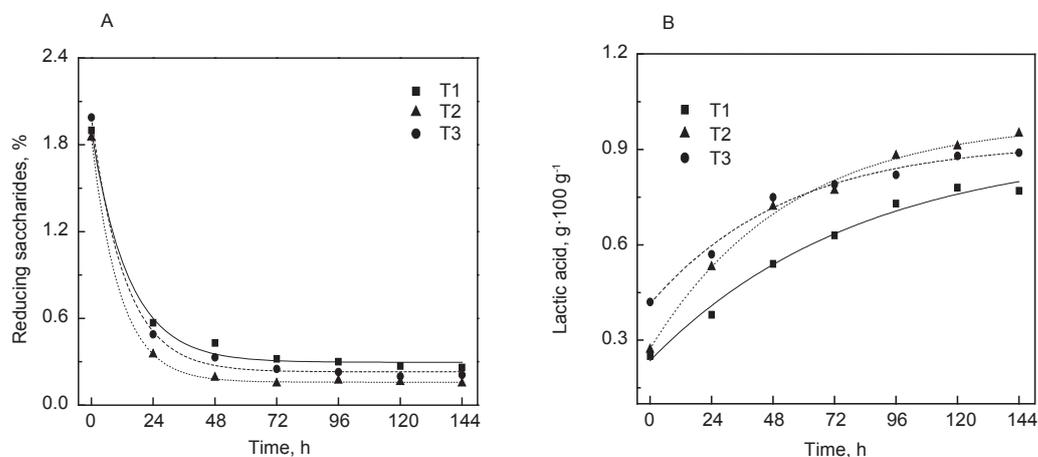


Fig. 2. Reducing saccharides decrease (A) and lactic acid production (B) during fermentation

al. [1999] during fermentation of tarhana samples with changed amount of added salt and yoghurt. The rapid increase in acidity minimizes the influence of spoilage bacteria. In the slowly acidified medium, lactic acid fermentation can be suppressed by butyric bacteria activity [Karovičová and Kohajdová 2003].

The content of reducing saccharides expressed as glucose (Fig. 2 A) varied from 1.85-1.99% at the beginning of the fermentation. Over a period of first 24 hours the content of saccharides rapidly declined to 0.35-0.57%. Further fermentation led to moderate decrease in reducing saccharides and after 144 hours achieved values in range 0.15-0.26%. Tamer et al. [2007] reported the content of reducing saccharides in various tarhana samples between 0.22-1.85% at the end of fermentation.

The organic acids, mainly lactic acid and acetic acid, produced by lactic acid bacteria are effective antimicrobial agents, and they reduce the pH in the food to prevent the growth of hazardous food microorganisms [Lee 1997]. The major organic acid formed in tarhana is lactic acid. The other acids formed such as acetic, propionic, citric and pyruvic constitute a very small amount of the total [Bozkurt and Gürbüz 2008]. The rate of lactic acid production during fermentation of tarhana is presented in Figure 2 B.

Lactic acid production in tarhana samples increased gradually from 0.25-0.42 g·100 g⁻¹ before fermentation to 0.77-0.95 g·100 g⁻¹ after 144 h. Increased amount of yoghurt and salt absence in tarhana doughs led to

higher production of lactic acid compared to the control sample. This could be explained that the increasing amount of yoghurt in tarhana formulation causes an increase in the total lactic acid bacteria count before and after fermentation, which also indicates higher lactic acid content in the product [Ozdemir et al. 2007].

Acetic acid in fermented products contributes to the aroma and prevents mould spoilage [Leroy and De Vuyst 2004]. Concentration of acetic acid (Fig. 3 A) increased during first 72 hours from 4.6 mg·100 g⁻¹ to 9.0-11.8 mg·100 g⁻¹. In later phases of fermentation (72-144 h) the content of acetic acid slightly decreased in case of sample T1 and T2, whereas in sample T3 concentration of acetic acid continued to increase (12.7 mg·100 g⁻¹ after 144 h). This could be explained that the acetate quantity depends mainly on the concentration of carbohydrates and the source of nitrogen, as well as the pH [Sroka and Tuszyński 2007].

The decrease in citric acid content during fermentation is attributed to its usage as a substrate in secondary reactions during fermentation. Citric acid is used up during metabolic activities of lactic acid bacteria and baker's yeast; consequently some metabolites, such as acetone and diacetyl, are formed [Caplice and Fitzgerald 1999]. In the present study, the concentration of citric acid (Fig. 3 B) gradually decreased from 76.5-59.0 mg·100 g⁻¹ in unfermented dough to 8.9-4.6 mg·100 g⁻¹ after 144 h fermentation process. The rate of citric acid degradation was comparable in all studied tarhana samples.

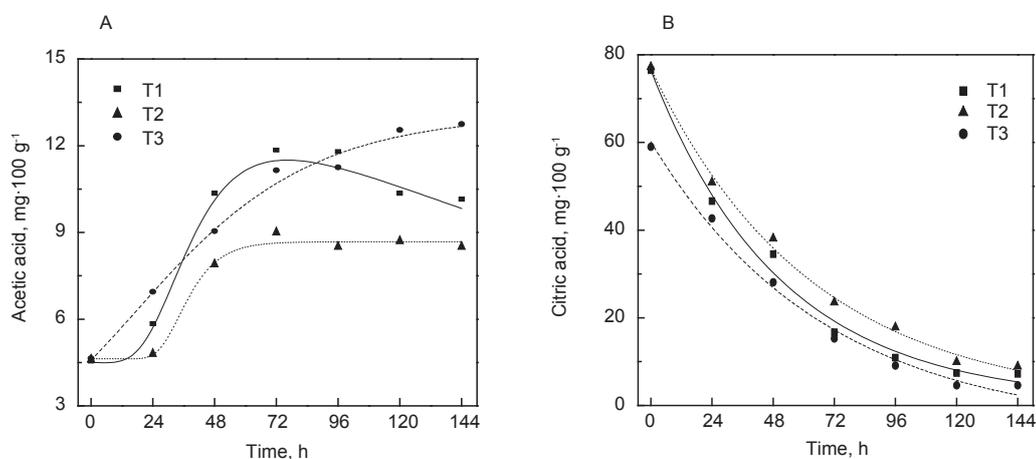


Fig. 3. Dependence between concentration of acetic acid (A) and citric acid (B) and time of tarhana fermentation

Table 2. Functional properties of tarhana

Sample	FC, cm ³ ·cm ⁻³	FS, min	WAC, cm ³ ·g ⁻¹	OAC, cm ³ ·g ⁻¹	EA, %
T1	0.21 ± 0.01 ^a	3.21 ± 0.10 ^a	0.65 ± 0.02 ^a	0.79 ± 0.01 ^a	93.13 ± 0.83 ^a
T2	0.19 ± 0.01 ^b	3.29 ± 0.04 ^a	0.73 ± 0.03 ^b	0.78 ± 0.01 ^a	93.96 ± 0.44 ^a
T3	0.12 ± 0.01 ^c	4.00 ± 0.13 ^b	0.70 ± 0.01 ^c	0.71 ± 0.02 ^b	90.30 ± 1.08 ^b

Values followed by different superscript letters in a column are significantly ($p = 0.05$) different from each other; T1 – control, T2 – without added salt, T3 – increased amount of yoghurt.

Fermentation, drying and different tarhana ingredients affect the functional properties of tarhana [Bilgiçli 2009, Çelik et al. 2005, Hayta et al. 2002]. Functional properties of tarhana samples are summarised in Table 2. Proteins in the dispersion resulted in lowering of the surface tension at the water-air interface, thus creating FC. Since FS is governed by the ability of the film formed around the entrapped air bubbles to remain intact without draining, it follows that stable foams can only be formed by the highly surface active solutes [Kaushal et al. 2012]. From this study resulted that addition of a higher proportion of yoghurt (sample T3) significantly reduced FC, whereas FS was increased. Tarhana contains milk proteins, mainly casein, and wheat proteins (for example glutenin and gliadins) originating from yoghurt and flour used in tarhana preparation [Hayta et al. 2002]. Some food

proteins are capable of forming good foams, and their capacity to form and keep stable foams depends on the type of protein, degree of denaturation, pH, temperature and processing methods [Hayta et al. 2002, Çelik et al. 2005]. Differences in FC and FS among the samples may reflect structural changes of proteins in tarhana during fermentation and drying [Çelik et al. 2005]. The decrease in FC of sample T3 can be explained by the proteolytic activity of present microorganisms, which lead to the weakness in the gas absorption property of proteins present in the tarhana formula [Çelik et al. 2005].

WAC is considered as an important property in viscous food such as sauces, dough and baked products [Hayta et al. 2002]. WAC is the ability to hold water against gravity. WAC comprised bound water, hydrodynamic water, capillary water and physically

Table 3. Sensory parameters of tarhana soups

Sample	Colour	Odour	Taste	Consistency	Overall acceptability
T1 – 72 h	6.57 ±0.21 ^a	6.23 ±0.25 ^d	6.36 ±0.23 ^a	6.43 ±0.12 ^d	6.20 ±0.20 ^e
T2 – 72 h	6.97 ±0.06 ^a	5.70 ±0.26 ^e	4.93 ±0.21 ^c	6.00 ±0.26 ^e	5.43 ±0.15 ^d
T3 – 72 h	5.77 ±0.25 ^c	6.00 ±0.10 ^f	5.63 ±0.15 ^d	6.40 ±0.17 ^f	6.33 ±0.25 ^e
T1 – 144 h	6.77 ±0.15 ^a	6.87 ±0.12 ^a	6.40 ±0.17 ^a	6.90 ±0.10 ^a	6.90 ±0.17 ^a
T2 – 144 h	6.60 ±0.10 ^a	6.10 ±0.30 ^b	5.20 ±0.20 ^b	6.33 ±0.25 ^b	6.00 ±0.30 ^b
T3 – 144 h	6.20 ±0.20 ^b	6.03 ±0.21 ^c	6.17 ±0.31 ^a	6.50 ±0.20 ^c	6.50 ±0.26 ^a

Superscript with different letters within each column denote statistically significant difference at level $p = 0.05$ from control (T1 – 144).

T1 – control, T2 – without added salt, T3 – increased amount of yoghurt.

entrapped water [Sridaran et al. 2012]. Control sample absorbed lowest amount of water ($0.65 \text{ cm}^3 \cdot \text{g}^{-1}$) while samples with changed recipe composition exhibit significantly different WAC (T2 $0.73 \text{ cm}^3 \cdot \text{g}^{-1}$ and T3 $0.70 \text{ cm}^3 \cdot \text{g}^{-1}$).

The OAC is an important functional property, as it helps to improve mouthfeel and retention of flavour [Ma et al. 2011]. Samples T1 and T2 had significantly different OAC (0.79 and $0.78 \text{ cm}^3 \cdot \text{g}^{-1}$) than tarhana T3 ($0.71 \text{ cm}^3 \cdot \text{g}^{-1}$). EA is the ability of the molecules to facilitate solubilization or dispersion of two immiscible liquids [Kohajdová et al. 2013]. From the measurements also resulted, that sample T3 (increased proportion of yoghurt) more significantly lower EA than other tarhana powder samples. The proteolytic activity of present microorganisms could be effective in this decrease of EA [Çelik et al. 2005].

Fermentation process is an important stage for the development of sensory profile [Erbaş et al. 2005]. Tarhana soup is made by fermented and dried tarhana dough, and it has acidic and sour taste with a yeasty flavour. The use of yoghurt (source of lactic acid bacteria) together with yeast plays an important role in developing distinctive tarhana taste and flavour [Çelik et al. 2010].

Sensory parameters of tarhana soup samples are presented in Table 3. Based on chemical analyses results (pH, acidity and organic acid content) it was evaluated and compared sensory parameters of samples fermented for 72 h and 144 h. Control represent sample T1 144 h and this sample was compared with the others. Samples with higher yoghurt proportion

(T3 72 h and 144 h) were presented with significantly lower values of colour. Odour and consistency evaluation showed, that studied samples differ significantly from the control sample. The highest overall acceptability reached control sample, while all samples fermented for 72 h showed significantly lower values of overall acceptability. This could be explained, that 72 h fermentation process is insufficient for development of various aroma and taste compounds.

CONCLUSION

Fermented foods contribute considerably to the human diet in many countries because fermentation is inexpensive, accessible and convenient preservation technology improving substantially the nutritional value and the organoleptic profile of food products [Kohajdová et al. 2007].

Fermentation of different tarhana samples (variation in tarhana recipe) presented in this study concluded in reduction of pH and content of reducing saccharides, increasing acidity of samples due to production of organic acid, predominantly lactic acid by lactic acid bacteria. It was found that using of the higher amount of yoghurt or salt absence in tarhana recipe resulted in higher lactic acid production. Furthermore, it was observed that increased proportion of yoghurt in tarhana negatively affected some functional properties (foaming capacity, oil absorption capacity and emulsifying activity), whereas foam stability and water absorption capacity were improved. Sensory characteristics evaluation showed, that tarhana samples fermented for 144 h

showed higher values of sensory attributes than samples fermented for 72 h and control sample reached the highest overall acceptability due to highest values of parameters such as taste, odour and consistency.

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