MICROBIOLOGICAL ATTRIBUTES OF INSTANT TARHANA DURING FERMENTATION AND DRYING

N. KARAGOZLU1, B. ERGONUL1* and C. KARAGOZLU2
1Celal Bayar University, Engineering Faculty, Food Engineering Department, Manisa, Turkey
2Ege University, Agricultural Faculty, Dairy Technology Department, Izmir, Turkey

Abstract


In this research, changes in chemical and microbiological attributes of traditional tarhana dough were investigated during an eight-day manufacturing period including fermentation and drying processes. Initial water content and pH value of tarhana dough were 47.97±1.76% and 4.97±0.08 respectively before fermentation process. At the end of the manufacturing process water content and pH value of instant tarhana were 5.8±0.66% and 4.19±0.04 (P<0.05). Total mesophylic aerobic bacteria, coliform bacteria, E.coli, mould, yeast and lactic acid bacteria counts of tarhana dough were 5.55±0.45 log cfu/g, <3 cfu/g, <3 cfu/g, <1 log cfu/g, 3.66±0.27 log cfu/g and 3.44±0.28 log cfu/g respectively, whereas these values were 5.38±0.23 log cfu/g, <3 cfu/g, <3 cfu/g, <1 log cfu/g, 5.38±0.3 log cfu/g and 4.75±0.12 log cfu/g at the end of the manufacturing process.

Key words: tarhana, fermentation, drying

Introduction

Tarhana is one of the widely consumed traditional fermented foods in Middle East countries and is of great importance in the diet of Turkish people. Tarhana is prepared by mixing yoghurt, wheat flour, yeast and a variety of cooked vegetables and spices (tomato, onion, salt, mint, paprika, tarhana herb, dill and basil) followed by fermentation for 1–7 days. Fermentation is usually carried out by yoghurt bacteria; Lactobacillus bulgaricus and Streptococcus thermophilus. After fermentation the mixture is sun dried and ground to a particle size of <1 mm (Ibanoglu et al., 1995). Instant tarhana is used to make tarhana soup by adding into water and boiling. Tarhana has an acidic and sour taste with a strong yeasty flavor and is also a good source of proteins and vitamins and therefore is used widely for feeding children and elderly people in the form of a thick soup (Turker and Elgun, 1995; Hammad and Fields, 1979) There are some other products similar to tarhana such as kishk (Tamime et al., 2000; Youssef, 1990), kushuk (Alnouri and Duitschaever, 1974), trahana (Economidou and Stein Kraus, 1983) and tahonya/talkuna (Siyamoglu, 1961). Methods for preparation of tarhana may vary from one place to another, but cereals and yoghurt are always the two major components. Since it has high nutritional properties, there is a growing commercial interest to produce tarhana soup in instant form (Ibanoglu and Ibanoglu, 1998).
Most of the tarhana consumed in Turkey is home-made and therefore sun-dried. However, there is a great commercial potential for the production of tarhana on an industrial scale using modern drying techniques (Ibanoglu and Maskan, 2002). The low pH and low moisture content make the tarhana a poor medium for pathogens and spoilage organisms; tarhana is not hygroscopic and it can be stored for 2 to 3 years without any sign of deterioration.

The scope of this work to monitor the chemical and microbiological changes during an 8-day tarhana manufacturing including 3-day fermentation, drying, grinding, sieving and packaging steps. Investigation of the microbiological changes would lead us to a conclusion on the chemical changes in tarhana during fermentation.

**Materials and Methods**

Manufacturing of tarhana was realized in Tarhana Plant of Menemen Research and Practice Farm of Agricultural Faculty, Ege University. Wheat flour (\textit{Triticum aestivum}, 13.20\% water, 12.80\% crude protein) was obtained from Tezcan Flour Ltd. Yoghurt was obtained from dairy plant of Agricultural Faculty, Ege University. The yoghurt used was fat set yoghurt (pH 4.7) made from cow’s milk having a dry matter of \%15.2, fat content of 3.0\% and 1.35 \% acidity (lactic acid). Tomato paste (30\% dry material) and paprika paste (32\% dry material) were obtained from Ege University. Also onion, salt, and tarhana herb used for tarhana preparation were purchased from local markets in Izmir. For the fermentation of tarhana dough, \textit{Saccharomyces cerevisiae} was used as starter culture and was obtained from Ege University Microbiology Laboratory.

**Production of Tarhana**

Method used for tarhana production was given as Figure 1. The composition of tarhana dough, based on total weight (wet basis), was as follows: wheat flour 900 g/kg; yoghurt, 300 g/kg; onion, 30 g/kg; tomato paste, 120 g/kg; salt 40 g/kg; yeast, 100 g/kg; paprika paste 20 g/kg; tarhana herb, basil and dill, 15 g/kg each. Tomato paste, paprika paste, onion, tarhana herb, basil and dill were smashed, blended and sieved. This mixture was pasteurized at 65 °C for 30 min and left for cooling down to 25°C. Then whole flour, salt, yoghurt and yeast were added into the mixture. They were kneaded in steel saucepan with a spoon to form tarhana dough. The dough was fermented at 30°C for 24 hours in an incubation room. Dough was left to fermentation for 1 day at room conditions. The following day, dough was cut into pieces. Dough pieces were left to be fermented for 2 more days. Next day, dough pieces were passed through crushing machine to obtain smaller pieces and then sieved. Powdered tarhana was left for drying process for 2 days at 35°C. After 2 days, second grinding was applied. Then, grinded tarhana was packaged.

Tarhana samples were taken on the 1st, 2nd, 3rd, 5th, 7th and 8th days of production period and changes in chemical (pH and moisture) and microbiological (total counts of mesophylic aerobic bacteria, coliform group bacteria, \textit{Echerichia coli}, yeast and mould counts and lactic acid bacteria count) attributes of tarhana were inspected. Tarhana was mixed well prior to sampling to ensure the homogenity of the samples. All the results are mean of two determinations with three replicates.

**Chemical Analysis**

To monitor the fermentation and keep under control, pH value and water content of the samples were determined. The pH (potentiometric) of the samples was measured with a pH-meter (WTW GmbH and Co., Model 537, Weilheim-Germany) and the moisture content was measured by drying the samples at 130±1°C in air oven (AOAC, 1980).

**Microbiological Analysis**

For the enumerations of Total Mesophylic Aerobic Bacteria (TMAB), Total Yeast and Mould (TYM), Lactic Acid Bacteria (LAB), and Coliform Bacteria, samples of tarhana (10 g) were dispersed in 90 ml
Fig. 1. Manufacturing process flow diagram for tarhana
sterile Ringer’s solutions and appropriate decimal dilutions were prepared by using 1/4-strength Ringer’s solution (Merck, Darmstadt, Germany) under the aseptic conditions. Total count of TMAB was enumerated in pour-plates of Plate Count Agar (Oxoid, CM0325) after incubation at 37°C for 48 hours. Dichloran Rose Bengal Chloramphenicol Agar (Oxoid) was used for TYM enumeration and plates were incubated at 25°C for 5 days. Lactic acid bacteria were enumerated in pour plates of de Man, Rogosa and Sharpe Medium (MRS) (Oxoid) after anaerobic incubation at 32°C for 3 days (Harrigan & McCance, 1976). Coliform group bacteria were enumerated in Violet Red Bile Agar (Oxoid) after incubation at 37°C for 24 h.

Statistical Analysis

Research was repeated three times and results of the duplicated analysis were statistically evaluated by using SAS Statistical Software (Release 7.00, SAS Institute Inc., Cary, NC, USA). Results are presented as mean ± standard deviation. Microbiological results were analyzed after used logarithmic transform. Significances were evaluated with the analysis of variance, followed by Duncan’s Multiple Range Test (P<0.05).

Results and Discussion

Water contents and pH values of tarhana samples found during manufacturing are given as Table 1. Before fermentation, water content of tarhana was 47.97±1.76%. After adding baker yeast into dough and incubated for 24 hours, water content was decreased to 34.73±4.3%. On the second day of the incubation water content of tarhana dough did not change. A rapid decrease in water content of tarhana was observed on 7th day of the fermentation period. Because of drying process performed on 5th and 6th days, water content of the sample was decreased to 6.13±1.06% from 18.87±1.34%. Drying process was found statistically significant on the water content of the samples (P<0.05). After grinding tarhana, because of the increased surface area, water content continued to decrease. Final water content of tarhana was 5.8±0.66% at the end of the 8-day manufacturing period. In their investigation into using different flours in tarhana production, Kose and Sungu-Çagýndi (2002) reported that, final acidity of tarhana produced by using wheat flour was 1.5% on dry basis, whereas the final moisture content was 10.7%. Ekinci (2005) stated that the water content of tarhana dough was 70.0%, and at the end of the manufacturing process, final water content was 10.0%.

Initial pH of tarhana was found as 4.97±0.08. By adding baker yeast and starting the fermentation process, at the end of the first day pH declined to 3.16±0.07. 1st incubation process was found significant on the pH changes of tarhana dough (P<0.05). With the second day of the incubation, pH value of tarhana increased to 4.14±0.22 then remained without any

<table>
<thead>
<tr>
<th>Process Step</th>
<th>Water (%)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tarhana dough</td>
<td>47.97±1.76a</td>
<td>4.97±0.08c</td>
</tr>
<tr>
<td>1st Incubation (1st day)</td>
<td>34.73±4.3b</td>
<td>4.16±0.07a</td>
</tr>
<tr>
<td>2nd Incubation (2nd day)</td>
<td>34.60±0.53b</td>
<td>4.14±0.22b</td>
</tr>
<tr>
<td>3rd Incubation (3rd day)</td>
<td>26.83±1.02c</td>
<td>4.08±0.19b</td>
</tr>
<tr>
<td>5th day</td>
<td>18.87±1.34d</td>
<td>4.14±0.08b</td>
</tr>
<tr>
<td>Grinded tarhana (7th day)</td>
<td>6.13±1.06e</td>
<td>4.10±0.06b</td>
</tr>
<tr>
<td>Packaged tarhana (8th day)</td>
<td>5.8±0.66e</td>
<td>4.19±0.04b</td>
</tr>
</tbody>
</table>

* Values with different letters in the same column are statistically different (P<0.05)
Microbiological Attributes of Instant Tarhana during Fermentation and Drying

Table 2
Microbiological analysis results of the raw materials used for tarhana production (n=3)

<table>
<thead>
<tr>
<th>Raw Material</th>
<th>TMAB (log cfu/g)</th>
<th>Coliform Bacteria (cfu/g)</th>
<th>E. coli (cfu/g)</th>
<th>Mould Count log cfu/g</th>
<th>Yeast Count log cfu/g</th>
<th>LAB log cfu/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yoghurt</td>
<td>7.65±0.24*</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>5.36±0.09</td>
<td>6.34±0.11</td>
</tr>
<tr>
<td>Red Pepper Paste</td>
<td>1.26±0.24</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Tomato Paste</td>
<td>1.20±0.17</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Onion</td>
<td>1.10±0.17</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Tarhana Herb</td>
<td>1.91±0.41</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Wheat Flour</td>
<td>5.34±0.4</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>3.38±0.2</td>
<td>2.43±0.91</td>
</tr>
</tbody>
</table>

*Values with different letters in a same column are statistically different (P<0.05)

Table 3
Changes in microbiological counts of tarhana during manufacturing

<table>
<thead>
<tr>
<th>Process Step</th>
<th>TMAB log cfu/g</th>
<th>Coliform Bacteria (cfu/g)</th>
<th>E. coli (cfu/g)</th>
<th>Mould Count log cfu/g</th>
<th>Yeast Count log cfu/g</th>
<th>LAB log cfu/g</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tarhana dough</td>
<td>5.55±0.45**a,c,d</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>3.66±0.27&lt;sup&gt;d&lt;/sup&gt;</td>
<td>3.44±0.28&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>1st Incubation (1st day)</td>
<td>6.66±0.04&lt;sup&gt;a&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>5.36±0.09&lt;sup&gt;c&lt;/sup&gt;</td>
<td>6.34±0.11&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>2nd Incubation (2nd day)</td>
<td>6.94±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>7.28±0.05&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.12±0.56&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>3rd Incubation (3rd day)</td>
<td>5.54±0.23&lt;sup&gt;c,d&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>6.54±0.19&lt;sup&gt;b&lt;/sup&gt;</td>
<td>7.37±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>5th day</td>
<td>5.26±0.05&lt;sup&gt;d&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>5.16±1.01&lt;sup&gt;c&lt;/sup&gt;</td>
<td>6.42±0.38&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Dried tarhana (7th day)</td>
<td>5.83±0.21&lt;sup&gt;b&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>5.31±0.45&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.40±0.47&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Packaged tarhana (8th day)</td>
<td>5.38±0.23&lt;sup&gt;c,d&lt;/sup&gt;</td>
<td>&lt;3</td>
<td>&lt;3</td>
<td>&lt;1</td>
<td>5.38±0.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.75±0.12&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

* Values with different letters in a same column are statistically different (P<0.05)
**Standard deviation

Ekinci (2005) stated that the initial pH value of tarhana dough before beginning of fermentation was 4.6±0.01 and as a result of 4-day fermentation pH value was declined to 4.0±0.01. In this period acidity was increased to 22.7±0.2 from 7.8±0.2. Ekinci reported that fermentation period was found statistically significant on the changes of pH value of tarhana dough (P<0.05).

As a result of their research, Erbas et al. (2006) reported that the pH value of tarhana dough was 4.63 before fermentation process. At the end of the first
day of the fermentation process pH value of dough were 4.24, whereas this value was 4.12 and 4.08 for the second and the third days of the fermentation (Erbas et al., 2006).

Initial pH content of tarhana dough was 4.97±0.08 before fermentation, whereas acidity of the sample increased to 11.93±1.65 from 7.65±0.48.

According to the research results of Ibanoglu et al. (1999) final moisture content of tarhana after manufacturing process was 7.7±0.1. pH value of tarhana was found as 4.80±0.01 in the same study.

Microbiological characteristics of raw materials used for manufacturing of tarhana are given as Table 2. As seen in Table 2, in none of the raw materials coliform group bacteria and *E. coli* were not detected. In all of the materials mould counts were <1 log cfu/g.

Changes in microbiological counts of tarhana dough during manufacturing process are given at Table 3. At the beginning of the fermentation, counts of TMAB, mould, yeast and LAB were found as 5.55±0.45, <1, 3.66±0.27 and 3.44±0.28 log cfu/g. Coliform group bacteria and *E. coli* were not detected in tarhana dough, On the first day of the fermentation period, TMAB count was increased to the level of 6.66±0.04 log cfu/g, whereas the yeast and LAB counts of dough were 5.36±0.09 and 6.34±0.11 log cfu/g. Yeast count and LAB count of dough were increased approximately 2 and 3 log units respectively. Increase in TMAB, yeast and LAB counts of tarhana dough on the first day of the fermentation was found statistically significant (*P*<0.05). In following day, TMAB count showed a little increase (*P*>0.05) and reached to 6.94±0.02 log cfu/g. Yeast and LAB counts of dough were 7.28±0.05 and 7.12±0.56 log cfu/g respectively. However, on the third day of the fermentation period, with an increase in the acid content of the dough, TMAB count of tarhana was declined to 5.54±0.23 log cfu/g. This decrease in TMAB count was found statistically significant (*P*<0.05). Similar pattern was seen for the yeast count. Yeast count of the sample was decreased to 6.54±0.19 log cfu/g (*P*<0.05). On the other hand, LAB count continued to increase. LAB count of tarhana was 7.37±0.11 log cfu/g on the third day of the fermentation. This decrease in LAB count was not found statistically significant (*P*>0.05). On the fifth day of manufacturing process, TMAB, Yeast and LAB counts of tarhana were all decreased and found as 5.26±0.05, 5.16±1.01 and 6.42±0.38 log cfu/g respectively. It is thought that this decrease was due to the decrease in pH value of tarhana. On the sixth and seventh day of the manufacturing period tarhana dough was left to final drying at 35°C. At this period, water content of dough was decreased to 6.13±1.06% from 18.87±1.34%. Due to loss of water from dough and decrease in water activity, TMAB count of dough was decreased to 4.40±0.47 log cfu/g. Yeast count of the sample was 5.31±0.45 log cfu/g. But, increase in yeast count of dough was not found statistically significant (*P*<0.05). On the eight day of the process, tarhana was packaged and the microbiological analysis results for TMAB, yeast and LAB counts were 5.38±0.23, 5.38±0.3 and 4.75±0.12 log cfu/g respectively. If compared with the seventh days results, the increase in LAB counts of the sample was not found statistically significant (*P*>0.05).

Ibanoglu et al. (1999) reported that initial TMAB, yeast and LAB counts of tarhana dough were 7.72, 6.92 and 7.82 log cfu/g respectively. These findings are higher than the results of our microbiological analysis results. On the first day of the fermentation period, Ibanoglu et al. (1999) found that the TMAB count was 8.43 log cfu/g. LAB and yeast counts were 7.98 and 7.45 log cfu/g. At the end of the fermentation period, these values were 7.65, 8.34 and 8.60 respectively. Also fermentation period was found statistically significant on the changes of microbiological counts of tarhana (*P*<0.05). Coliform bacteria and *E. coli* were not detected in any of the manufacturing steps. Also, no mould growth was seen.

**Conclusion**

Since, tarhana is a widely consumed traditional food especially in Eastern countries, it is very important to assure food safety measures in terms of microbiology. Tarhana is a dried food and the major microbiological risk is mould growth in the product. Traditional tarhana is sun-dried and contaminations during
drying cause an increase in microbiological counts of the product. Excessive amount of mould in the product may lead to mycotoxin synthesis. It is known that detoxification of a mycotoxin contaminated food product is impossible. So, preventive measures should be taken during fermentation and drying steps.

To assure a safe product, an affective HACCP plan should be implemented by the quality assurance department of the production plant and a controlled production programme should be adopted by taking microbiological risks into consideration.

References


