A review on traditional Turkish fermented non-alcoholic beverages: Microbiota, fermentation process and quality characteristics

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**A B S T R A C T**

Shalgam juice, hardaliye, boza, ayran (yoghurt drink) and kefir are the most known traditional Turkish fermented non-alcoholic beverages. The first three are obtained from vegetables, fruits and cereals, and the last two ones are made of milk. Shalgam juice, hardaliye and ayran are produced by lactic acid fermentation. Their microbiota is mainly composed of lactic acid bacteria (LAB). *Lactobacillus plantarum*, *Lactobacillus brevis* and *Lactobacillus paracasei* subsp. *paracasei* in shalgam fermentation and *L. paracasei* subsp. *paracasei* and *Lactobacillus casei* subsp. *pseudoplantarum* in hardaliye fermentation are predominant. Ayran is traditionally prepared by mixing yoghurt with water and salt. Yoghurt starter cultures are used in industrial ayran production. On the other hand, both alcohol and lactic acid fermentation occur in boza and kefir. Boza is prepared by using a mixture of maize, wheat and rice or their flours and water. Generally previously produced boza or sourdough/yoghurt are used as starter culture which is rich in *Lactobacillus* spp. and yeasts. Kefir is prepared by inoculation of raw milk with kefir grains which consists of different species of yeasts, LAB, acetic acid bacteria in a protein and polysaccharide matrix. The microbiota of boza and kefir is affected from raw materials, the origin and the production methods.

In this review, physicochemical properties, manufacturing technologies, microbiota and shelf life and spoilage of traditional fermented beverages were summarized along with how fermentation conditions could affect rheological properties of end product which are important during processing and storage.

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1. Introduction

Fermented food products play an important role in human diet around the world due to their health benefits. Fermentation is one of the oldest and most economical methods used in food preservation. The earliest records indicate that human were intaking ‘soured milks’ as long as 2000 years ago (Naidu et al., 1999). The beneficiary health effect of fermented milk products on humans was popularized by Elie Metchnikoff, who was a Nobel laureate for his works on concept of probiotics (Klaenhammer, 2007). The history of fermentation and modern fermented foods was well summarized by Hutkins (2006). In addition, fermentation enhances mineral bioavailability and the digestibility of proteins and carbohydrates and improves organoleptic qualities of the product (Hancioglu and Karapinar, 1997; Reddy and Pierson, 1994).

Lactic acid bacteria (LAB) have been used in various fermented foods. The preservative and health benefits of such traditional foods have been known for a long time (Hutkins, 2006; Klaenhammer, 2007; Naidu et al., 1999). The combination of this ancient method of bio-preservation with the current biotechnology tools should result in controlled fermentation processes and the selection of starter cultures to increase the consumption of fresh-like vegetables and fruits (McFeeters, 2004) and milk and milk-related components.

This review aimed to evaluate the microbiota of shalgam drink, hardaliye, boza, ayran and kefir known as traditional Turkish fermented non-alcoholic beverages in terms of their manufacturing technology. Quality characteristics and their affecting factors were summarized along with the spoilage of these beverages. Rheological characteristics of the beverages as a means of quality parameters and how they affected by the fermentation process were discussed for the first time in the literature.

2. Fermented non-alcoholic lactic acid beverages

The trends towards natural (minimally processed or without additives), highly nutritional value, health-promoting and flavor rich foods and beverages have been increased with consciousness of consumers. In this context, traditional Turkish fermented non-alcoholic beverages have been taking great attention from researchers and consumers recently due to their probiotic characteristics. The physicochemical properties, manufacturing technology, microbiota and spoilage of shalgam juice, hardaliye, boza, ayran and kefir were reviewed in this section, respectively.
2.1. Shalgam juice (fermented black carrot juice)

Shalgam is a red colored, cloudy and sour soft beverage which is produced by lactic acid fermentation of a mixture of turnips, black carrot bulgur (broken wheat) flour, salt and water. It is widely consumed in the cities of Adana, Hatay and Icel (the Mediterranean region of Turkey). In recent years, it has become popular in metropolises such as Istanbul, Ankara and Izmir (Tangüler and Erten, 2012a).

The composition of the juice has been reported by several authors as total dry matter (2.0–4.0%), protein (0.09–0.018%), salt (1.1–2.2%), lactic acid (0.578–8.05 g/L), ash (1.46–2.06%), and ethanol (0.79–6.41%). The titratable acidity is 1.06–9.1 g/L, and the pH varies between 3.15 and 4.25 (Arici, 2004; Canbas and Deryaoglu, 1993; Canbas and Fenercioglu, 1984; Özdeştan and Uren, 2010a; Özhan-Ozer, 2009; Tangüler and Erten, 2012b). The presence of sugar is not expected in the juice due to the fermentation (Erten et al., 2008).

Shalgam juice is a nutritional beverage due to high mineral, vitamin, amino acid and polyphenol contents (Erten et al., 2008; Inceday et al., 2008). It has been reported to contain anthocyanin as cyanidin-3-glycoside (88.3–114.1 mg/L) (Canbas, 1991). This compound is extracted from black carrot to shalgam juice during processing (Kammerer et al., 2004). The anthocyanin content of homemade shalgam juice (83.19 mg/L) was higher than commercial products (41.89–48.40 mg/L) (Ercelbi and Özkanli, 2010). Yilmaz-Ersan and Turan (2012) investigated the mineral contents of shalgam juices collected from domestic markets in Bursa, Turkey and reported that, sodium, potassium, calcium, magnesium, and phosphorus were the major elements, with heavy metals about 1 mg/L.

2.1.1. Manufacturing technology of shalgam juice

Shalgam juice is a homemade product; however, commercial manufacturing has increased recently (Canbas and Fenercioglu, 1984; Erten et al., 2008). There are differences in quality and stability due to variations in recipes and manufacturing techniques (Erten et al., 2008). There are two main processing methods for commercial production which are the traditional and the direct methods. The traditional method has two distinct fermentation stages: sourdough fermentation (first fermentation) and carrot fermentation (second fermentation). On the other hand, the direct method has only second fermentation (Erginkaya and Hammes, 1992; Erten et al., 2008).

In the traditional method, sourdough fermentation is performed by adding sourdough, salt and water to bulgur flour. Sourdough is obtained by using baker’s yeast dough fermented at room temperature for a few hours or overnight (Erten et al., 2008). The mixture is left to ferment at an ambient temperature for 3–5 days. The aim of sourdough fermentation is to increase the numbers of LAB and yeasts (Tangüler and Erten, 2012a). After the first fermentation, the mixture is extracted with water. Salt, sliced black carrot and sliced turnip are added to the extract in a tank for carrot fermentation (second fermentation). The mixture is then fermented for 3–10 days depending on the temperature (Canbas and Fenercioglu, 1984; Erten et al., 2008). After the second fermentation, a red colored, cloudy and sour shalgam juice is obtained by filtration and packaged in sealed bottles and plastic containers (Canbas and Fenercioglu, 1984; Erten et al., 2008).

In direct method, the fermentation tank is filled with the chopped black carrots, salt, sliced turnip, bakers’ yeast (Saccharomyces cerevisiae) or sourdough and adequate water. Fermentation occurs at ambient temperatures in between 10 °C and 35 °C for 3–5 days (Erten et al., 2008).

Even though these techniques are widely used in shalgam juice production, Cankurt et al. (2010) suggested an alternative method to decrease the fermentation period. They was used instead of water and the direct method was conducted. Thus, the fermentation period was shortened up to 80% (Cankurt et al., 2010).

2.1.2. Microbiota of shalgam juice

The microbiota of shalgam juice is mainly composed of LAB that belong to the genus Lactobacillus (89.63%) and followed by Leuconostoc (9.63%) and Pediococcus (0.74%). The total LAB level at the end of the first fermentation was reported within the range of 7.10–8.90 log cfu/g (Günes, 2008; Utus, 2008). The LAB level has increased at the first days of second fermentation than it gradually decreased to 6.02–8.23 log cfu/ml (Tangüler and Erten, 2012a,b; Utus, 2008). The high populations of LAB at the beginning of the fermentation were probably due to the raw materials, especially sourdough flora (Tangüler and Erten, 2012a).

Among the LAB, Lactobacillus plantarum, Lactobacillus brevis and Lactobacillus paracasei subsp. paracasei were predominant (Erginkaya and Hammes, 1992; Tangüler and Erten, 2012a,b). The occurrences of L. plantarum, L. paracasei subsp. paracasei, L. brevis, Lactobacillus buchneri and Pediococcus pentosaceus were reported at all stages of fermentation depending on the shalgam samples. L. plantarum, L. paracasei subsp. paracasei, Lactobacillus delbrueckii subsp. delbrueckii, P. pentosaceus, L. brevis and L. buchneri were isolated from first (dough) fermentation (Tangüler and Erten, 2012a,b,c). On the other hand, L. delbrueckii subsp. delbrueckii, Leuconostoc mesenteroides subsp. cremoris, L. mesenteroides subsp. mesenteroides and P. pentosaceus were only detected at the beginning of second fermentation (Tangüler and Erten, 2012a,b).

Besides LAB, yeasts that come in with the sourdough play a role in fermentation stages. However, the level of Saccharomyces and non-Saccharomyces yeasts is in lower level than LAB and their contribution to fermentation has not been clarified (Tangüler and Erten, 2012a).

The raw materials of shalgam juice contain sugars which can be fermented by LAB and yeasts. Among them, black carrot is considered as the main raw material and rich source of sugars (5.12–6.45 g/100 g) (Canbas and Deryaoglu, 1993). The main soluble fermentable sugars of black carrot are sucrose (1.20–3.31 g/100 g), glucose (1.10–5.60 g/100 g) and fructose (1.00–34.36 g/100 g) (Kammerer et al., 2004). Turnip and bulgur flour are the other sugar sources for LAB. Total sugar content of turnip is 3.80 g/100 g (USDA, 2011). It contains glucose (1.41 g/100 g), fructose (1.10 g/100 g) and sucrose in lower level (Rodríguez-Sevilla et al., 1999). The total sugar content of bulgur is 0.41 g/100 g (USDA, 2011).

Shalgam juice contains both homofermentative and heterofermentative LAB, Saccharomyces and non-Saccharomyces yeasts (Tangüler and Erten, 2012a). Depending on microbiota, the main products of fermentation are lactic acid (5.18–8.05 g/L), acetic acid (0.57–0.83 g/L), ethanol (in lower level) and volatile aroma compounds including carboxyl compounds, volatile acids, higher alcohols, esters, terpenols, norisoprenoids, lactones, and volatile phenols (Canbas and Deryaoglu, 1993; Tangüler and Erten, 2012a).

2.1.3. Shelf life and safety of shalgam juice

The shelf life of shalgam juice is 3 months at 4 °C in a sealed container. Pasteurization and/or the addition of preservatives can extend the shelf life up to 1–2 years, but the sensory properties are adversely affected due to cooked flavor of carrot (Canbas and Fenercioglu, 1984; Erten et al., 2008). Commercially, the shelf life is extended up to 1 year with pasteurization and the addition of benzoic acid and its salts (Tangüler and Erten, 2012a).

Shalgam juices should have a pH 3.3–3.8 (TS, 2003), in which case it is not spoiled by undesirable bacteria. Yeasts may grow in the acid environment and cause spoilage by film formation, color changes and off-flavor. Recently, Candida krasei, Candida pelliculosa and Candida lipolytica were reported as the causative yeasts in the spoiled shalgam juices (Özhan-Ozer, 2009).

In two different studies, the survival of Escherichia coli (Özhan and Coksoyler, 2005) and Salmonella Typhimurium (Tosun and Gönül, 2003) in shalgam juices was investigated. During storage, the growth of E. coli at an ambient temperature within 19 h (Ôzhan and
Coksoyler, 2005) and acid-adapted and non-acid-adapted Salmonella Typhimurium at 4 °C and 20 °C within 24 h was inhibited due to low pH (Tosun and Gönül, 2003).

The total biogenic amine contents of shalgam juices were determined as in the range of 26.7–134.3 mg/L. In peppered and non-peppered samples, putrescine, cadaverine, histamine and tyramine were determined and putrescine had the highest levels (5–42.3 mg/L) and followed by tyramine (3.8–41.0 mg/L) (Özdestan and Üren, 2010a). These amines could be produced by the LAB associated with the fermentation, but the levels found may not be high enough to cause adverse effects on consumers (EFSA, 2011; Silla-Santos, 2001).

2.2. Hardaliye

Hardaliye is a kind of grape based non-alcoholic traditional beverage. It is one of the popular beverages consumed in the Thrace in the Marmara region of Turkey (Anonymous, 2011). Due to the LAB flora of hardaliye; it has been classified as non-dairy probiotic beverage (Prado et al., 2008).

Limited studies about hardaliye have been presented in the literature due to fact that it is a local fermented drink in Turkey. The pH of hardaliye was reported to be in the range of 3.21 and 3.97 (Arici and Coskun, 2001; Güven and Aksoy, 2009) whereas the pH values of clover or ginger added hardaliye samples were slightly higher at 3.94 and 4.11, respectively. The ginger added hardaliye had the lowest titratable acidity value (4.14%). Similar results for clover added and traditionally produced hardaliye in terms of titratable acidity, brix and reducing sugar were determined by Güven and Aksoy (2009).

The color intensities of hardaliye change in a wide range depending on grape varieties and production methods (Arici and Coskun, 2001; Güven and Aksoy, 2009).

2.2.1. Manufacturing technology of hardaliye

Hardaliye is mostly manufactured homemade by the traditional method. Red grape (Papazkarasi, Alfonso or Cardinal) or grape juice and crushed black mustard seed and cherry leaf are used for hardaliye production. Washed red grapes and mustard seeds are pressed separately till the rupture of their crusts. The ruptured crust of grapes gives the dark color to final product depending on grape varieties. Pressed grapes and cherry leaves are placed into a barrel and 0.2% pressed mustard seeds and/or 0.1% of benzoic acid are added. The barrels are closed and incubated at room temperature for 5–10 days. After incubation, the mixture is filtered and kept at cold (Arici and Coskun, 2001).

2.2.2. Microbiota of hardaliye

There is only one study on the microbiology of hardaliye fermentation in the literature. The numbers of LAB were reported within the range of $1 \times 10^{2}–4 \times 10^{5}$ cfu/mL. L. paracasei subsp. paracasei and L. casei subsp. pseudoplanturn were predominantly followed by L. pontis, L. brevis, L. acetotOLERANS, L. sanfranciscensis (formerly known as Lactobacillus sanfrancisco), and L. vaccinostercus (Arici and Coskun, 2001).

2.2.3. Shelf life and safety of hardaliye

Due to the low pH and antimicrobial components of aged hardaliye, the microbial counts were determined as in the range of 26.7–134.3 mg/L. In peppered and non-peppered samples, putrescine, cadaverine, histamine and tyramine were determined and putrescine had the highest levels (5–42.3 mg/L) and followed by tyramine (3.8–41.0 mg/L) (ÖZDESTAN and ÜREN, 2010a). These amines could be produced by the LAB associated with the fermentation, but the levels found may not be high enough to cause adverse effects on consumers (EFSA, 2011; Silla-Santos, 2001).

2.3. Boza

Boza is a traditional Turkish non-alcoholic fermented beverage produced from millet, maize, wheat, or rice semolina or flour by yeast and lactic acid fermentation. It is a viscous liquid with a pale yellow color and sweet or sour taste. It is widely consumed in Turkey, Bulgaria and some other countries of the Balkan Peninsula due to its pleasant taste, flavor and its nutritional properties (Akpinar-Bayizit et al., 2010; Gotcheva et al., 2001; Yegin and Fernandez-Lahore, 2012).

The compositional variations in boza samples result from the utilization of different types and amounts of cereals and cereal products as a raw material, and uncontrolled fermentation conditions (Akpinar-Bayizit et al., 2010; Gotcheva et al., 2000, 2001). The total dry matter, protein, total sugar, ash, titratable acidity, pH and ethyl alcohol contents of boza samples were varied from 5.57% to 29.82%, 0.27% to 2.75%, 10.64% to 22.59%, 0.02 to 0.17%, 0.15 to 0.5%, 3.16 to 4.63 and ND to 0.39%, respectively (Gotcheva et al., 2000; Köse and Yücel, 2003; Meric, 2010; Uylaser et al., 1998; Uysal et al., 2009; Yegin and Üren, 2008).

Akpinar-Bayizit et al. (2010) investigated the effect of raw materials such as rice, millet and wheat on the chemical composition of boza and reported that the highest total titratable acidity value was in wheat boza (0.61 ± 0.07%) due to the high fermentable carbohydrate content of wheat compared to other raw materials, whereas millet boza had the lowest titratable acidity (0.32 ± 0.04%). The pH varied in between 3.43 ± 0.08 and 3.86 ± 0.17 depending on the raw material. The organic acid profiles and the alcohol contents were also affected by the choice of raw materials. Oxalic, lactic, pyruvic, and acetic acid were found in all samples while malic acid was found only in rice boza. Citric acid was detected in rice and maize boza whereas orotic acid was found in rice, maize and millet boza (Akpinar-Bayizit et al., 2010).

Fermentation time and temperature are also important factors affecting the physicochemical properties. The extended fermentation time results in lower pH and higher titratable acidity levels. In a study, after 24 hour fermentation the pH decreased from 6.13 to 3.48 whereas the total titratable acidity and the alcohol content increased from 0.02% to 0.27% and from 0.02% to 0.79%, respectively (Hancioglu and Karapinar, 1997). Köse and Yücel (2003) reported that the initial pH of unfermented boza was in between 4.1 and 6.7 and it could decrease to 4.0 or below during fermentation.

2.3.1. Manufacturing technology of boza

There are six main stages in the boza production which are preparation of raw materials, boiling, filtration and cooling, sugar addition, fermentation and bottling (Arici and Daglioglu, 2002). The selection of raw materials is an important stage, which affects the degree of fermentability, viscosity and dry matter content (Akpinar-Bayizit et al., 2010; Gotcheva et al., 2001). In the preparation step, cereal grains are cleaned and milled to the size of 300–800 μm (Arici and Daglioglu, 2002; Yegin and Üren, 2008). Water is added at the 1:5 ratio of cereals:water to milled grains and the mixture is boiled for 1–2 h depending on boiling temperature and raw material. Boiling process is carried out until homogenous pulp formation is observed (Arici and Daglioglu, 2002). The mixture is filtered to remove bran, hull and other foreign materials (Arici and Daglioglu, 2002; Yegin and Üren, 2008). After that, it is cooled by stirring to prevent the formation of a thin layer on the surface. Sugar/saccharose powder (20–25%) is added to mixture as a substrate for LAB and yeasts (Arici and Daglioglu, 2002; Yegin and Fernandez-Lahore, 2012). Previouslyhardaliye samples containing white (Brassica alba (L.) Boiss) or black (Brassica nigra (L.) Koch) mustard seeds. On the other hand lower yeast and mold counts were observed in hardaliye samples containing black mustard seeds (Coskun and Arici, 2011).
fermented boza can be used as starter culture. Fermentation occurs at 30 °C for 24 h (Yegin and Uren, 2008). Two types of fermentation are observed in boza which are lactic acid fermentation by LAB and alcohol fermentation by yeasts. LAB produce antimicrobial substances and increase acidity which provides preservative effect (Arici and Daglioglu, 2002). Antimicrobial substances produced by LAB such as lactic acid are considered as one of the factors of the unique taste of boza (Kabak and Dobson, 2011). Metabolites of alcohol fermentation affect the odor and mouthfeel of boza (Yegin and Fernandez-Lahore, 2012). After fermentation stage, boza is cooled and bottled in plastic or glass containers (Arici and Daglioglu, 2002).

2.3.2. Microbiota of boza

The microbiological properties, LAB and yeasts isolated from boza samples were shown in Tables 1 and 2, respectively. As seen in Table 1, the LAB and yeast total counts found in boza vary within the range of 2.94 × 10⁷–4.6 × 10⁸ cfu/mL and 2.24 × 10⁵–8.40 × 10⁷ cfu/mL, respectively. It has been reported that after 24 h fermentation the number of LAB and yeasts increased from 7.6 × 10⁶ to 4.6 × 10⁸ cfu/mL and 2.25 × 10⁵ to 8.10 × 10⁶ cfu/mL, respectively (Hancioglu and Karapinar, 1997).

Hancioglu and Karapinar (1997) isolated 77 LAB and 70 yeast strains. Among the LAB, Leuconostoc paramesenteroides (25.6%) was predominant, followed by L. sanfrancisco (21.9%) and L. mesenteroides subsp. mesenteroides (18.6%). On the other hand, Gotcheva et al. (2000) reported that L. plantarum (24% of LAB), Lactobacillus acidophilus (23% of LAB) and L. fermentum (19% of LAB) were predominantly isolated from Bulgarian boza. L. plantarum was also frequently isolated from Turkish boza samples by Kivanc et al. (2011) (Table 2).

Hancioglu and Karapinar (1997) have reported that 83% of yeast isolates were S. uvarum and the remainder were S. cerevisiae. Gökmen et al. (2000) isolated 50 yeasts from boza samples obtained from local markets and Candida spp. was predominant, followed by Saccharomyces, Trichosporon, Torulospora and Rhodotorula. In Bulgarian boza, S. cerevisiae, C. tropicalis and C. glabrata comprised the majority of yeasts (Gotcheva et al., 2001). Recently, Pichia fermentans was reported as the most representative yeasts in Turkish boza (Caputo et al., 2012). Diversity in fermentation flora and variations in numbers of LAB and yeast from the different samples could be due to different raw materials, production processes and storage conditions (Botes et al., 2007; Gotcheva et al., 2001).

Bacteriocin and antimicrobial metabolite producing LAB such as L. plantarum (Todorov and Dicks, 2006; Todorov, 2010), L. pentosus (Todorov and Dicks, 2006), L. rhamnosus (Todorov and Dicks, 2006), L. paracasei (Todorov and Dicks, 2006), Lactobacillus acidophilus (Todorov et al., 2008), Lactococcus lactis subsp. lactis (Akkoe et al., 2011; Tuncer et al., 2008), Leuconostoc lactis (Todorov, 2010), and P. pentosaceus (Todorov and Dicks, 2005) were isolated from boza. Boza is also regarded as a rich source of probiotic bacteria. Probiotic properties of L. plantarum, L. paracasei, L. rhamnosus and L. pentosus were evaluated by Todorov et al. (2008).

The selection of cultures is important to obtain product with desirable quality and stability (Yegin and Fernandez-Lahore, 2012). Zorba et al. (2003) investigated the appropriate starter culture combinations using sensory evaluation. S. cerevisiae/L. mesenteroides subsp. mesenteroides/L. confusus (Weissella confusa) combination was suggested as a starter culture mixture to obtain desirable sensory characteristics.

Further work is needed to improve the quality and stability of boza. By using LAB and yeast isolates as starter cultures, controlled fermentation studies can be carried out. The selection of proper strains with probiotic and antimicrobial properties also enhances the functional properties of boza.

2.3.3. Shelf life and safety of boza

The shelf life of boza is fairly short, up to 15 days. Boza is not appropriate for storage below 10 °C. Boza is acceptable for consumption at every stage of the fermentation until pH drops to about 3.5 (Gotcheva et al., 2001).

Total aerobic bacteria count was detected within the range of 1.53 × 10⁹–2.30 × 10⁶ cfu/mL (Table 1). The occurrence of coliforms and molds was reported in some of the samples; however E. coli, Salmonella, Staphylococcus aureus and Bacillus cereus were not detected. The occurrence of opportunistic human pathogens such as Candida inopsicica, Rhodotorula mucilaginosa and Pichia norvegensis was reported by Botes et al. (2007). Aspergillus fumigatus, Penicillium chrysogenum, Fusarium oxysporum, Acremonium spp., Geotrichum candidum and Geotrichum capitatum were isolated from boza. Although the ochratoxigenic molds have not been reported in the boza, ochratoxin A was determined in two of five samples. The source of ochratoxin A might be the raw materials (Yusal et al., 2009).

Due the activity of fermentation microorganisms, the occurrences of biogenic amines in boza were reported within the ranges 13–65 mg/kg and 1.67–101.14 mg/kg by Yegin and Uren (2008) and Cosansu (2008), respectively. Putrescine, spermidine and tyramine were detected in all boza samples. On the contrary, lower incidences for putrescine (33%) and tyramine (14%) were reported by Cosansu (2009). Among the bioamines in boza, tyramine (4.68–82.79 mg/kg) has the highest levels, which can cause adverse health effects for consumers who are taking classical MAOI drugs.

2.4. Ayran (yoghurt drink)

Ayran is a drinkable fermented milk product produced by the addition of water to yoghurt (homemade) or by the addition of Streptococcus thermophilus and Lactobacillus bulgaricus to standardized milk for fermentation (industrially produced) (TFC, 2009). Ayran is widely consumed in Turkey especially in summer season. The chemical composition of ayran depends on the type of milk used.

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**Table 1**
The microbiological properties of boza samples.

<table>
<thead>
<tr>
<th>Sample properties</th>
<th>Total aerobic bacteria (cfu/mL)</th>
<th>Lactic acid bacteria (cfu/mL)</th>
<th>Coliforms (cfu/mL)</th>
<th>Yeasts (cfu/mL)</th>
<th>Molds (cfu/mL)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laboratory prepared (Turkey)</td>
<td>4.60 × 10⁶</td>
<td>8.10 × 10⁶</td>
<td></td>
<td></td>
<td></td>
<td>Hancioglu and Karapinar (1997)</td>
</tr>
<tr>
<td>Local markets (Bulgaria)</td>
<td>2.40 × 10⁵–3.20 × 10⁶</td>
<td>6.00 × 10⁵–8.80 × 10⁷</td>
<td></td>
<td>2.60 × 10⁷–3.90 × 10⁷</td>
<td></td>
<td>Gotcheva et al. (2000)</td>
</tr>
<tr>
<td>Local markets (Turkey)</td>
<td>1.53 × 10⁵–5.90 × 10⁶</td>
<td>2.10 × 10⁵–2.90 × 10⁷</td>
<td>ND–110</td>
<td>4.70 × 10⁵–5.40 × 10⁶</td>
<td></td>
<td>Tuncer et al. (2008)</td>
</tr>
<tr>
<td>Laboratory prepared (stored at 20 °C for 10 days) (Turkey)</td>
<td>8.32 × 10⁵–3.09 × 10⁶</td>
<td>5.89 × 10⁵–4.47 × 10⁶</td>
<td>ND–4.47 × 10⁶</td>
<td>1.29 × 10⁶–6.17 × 10⁶</td>
<td></td>
<td>Yusal et al. (2009)</td>
</tr>
<tr>
<td>Local markets (Turkey)</td>
<td>3.90 × 10⁴</td>
<td>1.29 × 10⁵</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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* ND, not detected.
* Yeast-mold count.
* On MRS.
* on M17.
Table 2  
Lactic acid bacteria and yeasts isolated from boza 

<table>
<thead>
<tr>
<th>Genus</th>
<th>Species</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactobacillus</td>
<td>acidophilus</td>
<td>Gotcheva et al. (2000, 2001)</td>
</tr>
<tr>
<td></td>
<td>brevis</td>
<td>Gotcheva et al. (2000), Botes et al. (2007),</td>
</tr>
<tr>
<td></td>
<td>coryniformis</td>
<td>Kivanc et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>fermentum</td>
<td>Hancioglu and Karapinar (1997)</td>
</tr>
<tr>
<td></td>
<td>graminis</td>
<td>Hancioglu and Karapinar (1997), Gotcheva et al.</td>
</tr>
<tr>
<td></td>
<td>paracasei</td>
<td>(2000, 2001), Botes et al. (2007)</td>
</tr>
<tr>
<td></td>
<td>paracasei subsp. paracasei</td>
<td>Kivanc et al. (2011)</td>
</tr>
<tr>
<td></td>
<td>paraplanatum</td>
<td>Botes et al. (2007), Kivanc et al. (2011)</td>
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<td>pentosus</td>
<td>Botes et al. (2007), Todorov and Dicks (2006)</td>
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<td>plantarum</td>
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<td>rhamnosus</td>
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<td>Botes et al. (2007), Todorov and Dicks (2006),</td>
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<td>Lactococcus</td>
<td>lactis subsp. lactis</td>
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<td>Leuconostoc</td>
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<td></td>
<td>mesenteroides</td>
<td>Gotcheva et al. (2000)</td>
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<td>mesenteroides subsp. mesenteroides</td>
<td>Hancioglu and Karapinar (1997)</td>
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<td>paramesenteroides</td>
<td>Gotcheva et al. (2000)</td>
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<td></td>
<td>raffinolactis</td>
<td>Gotcheva et al. (2000)</td>
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<tr>
<td>Oenococcus</td>
<td>oeni (formerly known as Leuconostoc oenos)</td>
<td>Hancioglu and Karapinar (1997)</td>
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<td>Pediococcus</td>
<td>spp.</td>
<td>Kivanc et al. (2011)</td>
</tr>
<tr>
<td>Weissella</td>
<td>confusa (formerly known as Lactobacillus coprophilus and Lactobacillus confusus)</td>
<td>Todorov and Dicks (2005)</td>
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Yeasts  
Candida                  
Candida                  diversa                       Botes et al. (2007)                  
                              glabrata                     Gotcheva et al. (2000, 2001)                 
                              inconspicua                  Botes et al. (2007), Caputo et al. (2012)  
                              paraarbus                    Botes et al. (2007)                          
                              quercitrusa                  Caputo et al. (2012)                        
                              silvae                       Caputo et al. (2012)                        
                              tropicalis                   Gotcheva et al. (2000, 2001)               
Clavispora               
Coniochaeta              
Cryptococcus             
Cystofibobasidium        
Geotrichum               
Issatchenkia             
Pichia                   
Saccharomyces            
Torulaspora              
Trichosporon            

* Identification of microorganisms was carried out by examination of morphological and physiological characteristics by Hancioglu and Karapinar (1997), Gotcheva et al. (2000, 2001), Todorov and Dicks (2004, 2005) and by molecular methods by Botes et al. (2007), Caputo et al. (2012), Todorov and Dicks (2006), Todorov (2010) and Kivanc et al. (2011). The efficiency of fat removal and the dilution rate (Kabak and Dobson, 2011). The composition of ayran has been reported by several authors as the total dry matter (1.07–11%), protein (1.44–3.48%), salt (0.17–1.75%) and fat (0.1–3%). The titratable acidity is 0.4–1.73% and the pH varies between 3.44 and 4.44 (Gülmez et al., 2003; Kocak et al., 2006; Sanli et al., 2011; Sen and Küplülü, 2004; Patir et al., 2006; Tumucay-Özünlü and Kocak, 2010a).

Ayran is easily digestible and a highly valued drink with high content of vitamin and calcium. Indeed, it is possible to develop the functional properties of ayran. Kök-Tas and Güzel-Seydim (2010) have produced ayran by addition of inulin as prebiotic and L. acidophilus and Bifidobacterium spp. as probiotic strains. They have reported that the product had good taste, appearance and higher Lactobacillus bacteria count.

The main textural defects are low viscosity and serum separation (up to 30%). Serum separation occurs in fermented milk products due to the aggregation of casein micelles to particles and sedimentation of casein particles during storage. The presence of salt in ayran may cause more serum separation compared to other fermented milk beverages. Addition of increasing levels of salt and water was found to increase the serum separation in ayran (Köksoy and Kilic, 2003). Serum separation can be prevented by addition of hydrocolloid stabilizers such as pectin, guar gum and gelatin (Köksoy and Kilic, 2004) or addition of transglutaminase (Sanli et al., 2011).
significant effect of stabilizers on taste, odor, consistency and overall acceptability was determined (Köksoy and Kilic, 2004). Transglutaminase has been used to modify functional properties of food proteins. The addition of transglutaminase a level of 1 Tgase/g was no significant effect on flavor or chemical properties of ayran (Sanli et al., 2011).

Tamucay-Özünlü and Kocak (2010a) investigated the effects of different heat treatments of milk (at 75, 85 and 95 °C for 5 min) on the various properties of ayran. The acetaldehyde content, serum separation and viscosity were significantly affected. Heat treatment at 95 °C was recommended to obtain the highest viscosity and the least serum separation.

2.4.1. Manufacturing technology of ayran

Ayran is a fermented beverage which is traditionally prepared by blending yoghurt with water (30–50%) and salt (0.5–1%) (Köksoy and Kilic, 2003). Homemade ayran is produced daily and consumed fresh; because of that there is no need for individual packaging commercially when homemade ayran is sold in restaurants, buffets and pastries.

Industrial production of ayran can be carried out by two different methods. It can be produced either by the addition of water to yoghurt or the addition of water to milk first and then fermentation of diluted milk (Kocak et al., 2006; Kocak and Avsar, 2009). The processes for industrial production of ayran are outlined in Fig. 1. As a first stage raw milk is standardized in terms of fat (1.5% for full fat, 0.8% for half-fat, 0.15% for fat free). Standardized milk is diluted with water until 8% of total solid content is obtained in technique 1. In technique 2, water is not added; milk is homogenized and pasteurized in both techniques (Kocak and Avsar, 2009). By pasteurization, it is possible to obtain ayran with good microbiological quality (Kocak and Avsar, 2009; Sen and Küplülü, 2004). Pasteurized milk is inoculated with yoghurt starter cultures (S. thermophilus and L. delbrueckii subsp. bulgaricus) and incubated until a pH 4.2–4.4 is obtained. Then the fermented samples are cooled in order to end the fermentation and development of acidity (Köksoy and Kilic, 2003; Kocak and Avsar, 2009). Adequate water is added to fermented milk in technique 2, until the total solid content of fermented sample reaches to 8%. After that salt (0.5%) is added. Industrially produced ayran is bottled in a glass or polypropylene or polystyrene plastic containers when it is industrially produced in dairy plants (Sen and Küplülü, 2004).

Different manufacturing techniques affect acetaldehyde content but these differences are not detectable in sensory analysis (Kocak et al., 2006).

2.4.2. Microbiota of ayran

The microbiota of homemade ayran is similar to the microbiota of yoghurt which is used for its production. Yoghurt bacteria which are S. thermophilus and L. delbrueckii subsp. bulgaricus are used in the fermentation of milk in industrial production. The numbers of yoghurt bacteria in industrially produced ayran are higher than homemade ayran. Selection of starter culture is critical on rheological and texturial properties of ayran (Kocak and Avsar, 2009). A bitter flavor can develop as a result of the production of bitter peptides by some strains of L. delbrueckii subsp. bulgaricus used as starter cultures. During storage, starter cultures can continue to produce lactic acid, causing an objectionable sharp and acid taste (Ray, 2001). Phage sensitivity of starter cultures is an important issue in terms of selection of culture, as well. Phage infection of starter cultures can cause a negative impact on functionality of starter cultures (Soykut and Tunail, 2009). In a study, bacteriophages of S. thermophilus were isolated from 79 samples obtained from dairy plant. The numbers of phages ranged from 10^2 to 10^7 cfu/mL (Quiberoni et al., 2003).

There are several factors which could affect the content of yoghurt bacteria in ayran production (Akalin and Gönc, 1999). It has been reported that the pH can vary with the levels of L. delbrueckii subsp. bulgaricus and S. thermophilus. Depending on the increase of acidity, the level of L. delbrueckii subsp. bulgaricus has increased; the level of S. thermophilus has decreased due to the inhibition effect of acidity on S. thermophilus (Tamucay-Özünlü and Kocak, 2010b).

2.4.3. Shelf life and safety of ayran

The shelf life of ayran is limited to 10 to 15 days at 4 °C. It is not possible to apply any heat treatments or other processes for extending its shelf life because such processes would also decrease the number of yoghurt bacteria, which must be higher than 10^7 cfu/mL (TFC, 2009) according to the Turkish Food Codex (Kocak and Avsar, 2009).

The industrially produced ayran microbiota is more stable compared to the microbiota of homemade ayran. The risk of contamination is very high for homemade ayran and particularly Kluyveromyces spp. and Saccharomyces spp., are often present (Kocak and Avsar, 2009). The occurrence of Kluyveromyces lactis, S. cerevisiae and Geotrichum spp. in ayran has been reported at low levels. Microbiological properties of ayran have been shown in Table 3. Sen and Küplülü (2004) investigated microbiological and chemical properties of 35 homemade ayran samples. They reported that none of the samples were acceptable according to the regulations in terms of yeast count. They also indicated that 28.5% and 77.1% of samples did not meet the criteria of limit values for coliform bacteria count and mold count, respectively. Patir et al. (2006) also detected higher numbers of microorganism in homemade ayran.
samples compared to industrially produced samples. 88% of homemade ayran samples and 18% of industrially produced samples were contaminated with coliform bacteria greater than 10 MPN/mL. On the other hand, Tamucay-Özünülü and Kocak (2010b) observed no coliform bacteria in their laboratory prepared ayran samples and the yeast-mold level of all samples was below the limit value. In addition, Simsek et al. (2007) reported that E. coli O157:H7 was inhibited during ayran fermentation.

2.5. Kefir

Kefir is a viscous and self-carbonated beverage with a smooth, slightly foamy body and whitish color (Mistry, 2004; Yuksekdağ et al., 2004). It is originated in the Caucasus Mountains and produced by fermentation of cow, ewe, goat or other type of milk (Kabak and Dobson, 2011).

Physicochemical properties and quality of kefir are affected from the microbial quality of kefir grains, the grain to milk ratio, incubation time and temperature, sanitation conditions and storage temperature (Guzel-Seydim et al., 2010). The composition of kefir has been reported by several authors as the total dry matter (8.88–16.73%), protein (3.10–4.72) and fat contents (1.11–2.77) (Cetinkaya and Elal-Mus, 2012; Dinc, 2008; Ertekin and Guzel-Seydim, 2010; Sady et al., 2007; Uslu, 2010).

The type and amount of milk are important in terms of sensorial and textural properties of kefir (Garrote et al., 1998; Wszolek et al., 2001). On the other hand, Oner et al. (2010) found no differences on chemical and microbiological properties of kefir made from different types of milk. As a result of fermentation lactic acid, ethanol, carbon dioxide and other flavor compounds such as acetaldehyde, diacetyl and acetoin occur in typical kefir product. Among them, the ethanol content (0.035% to 2%) of kefir was determined in a wide range (Guzel-Seydim et al., 2010).

2.5.1. Manufacturing technology of kefir

Kefir can be made from any kind of milk including cow, goat, sheep, camel, buffalo milk or milk substitutes such as soy milk, rice milk and coconut milk (Irigoyen et al., 2005; Otles and Cagindi, 2003). In addition, milk can be pasteurized, unpasteurized, whole fat, low fat, skim and no fat. There are mainly two methods for manufacturing of kefir: traditional (authentic) and industrial (commercial) processing (Guzel-Seydim et al., 2010; Otles and Cagindi, 2003). The process for production of kefir by traditional method is outlined in Fig. 2. In the traditional method, kefir grains are directly added to the pasteurized and cooled milk and incubated with stirring for approximately 24 h at 25 °C. After fermentation, the grains are separated from the milk by filtering with a sterile sieve and can be dried at room temperature and kept at cold storage for the next inoculation. Kefir is stored at 4 °C for a time and then is ready for consumption. Industrial (commercial) process of kefir differs from the traditional methods. Lyophilized starter cultures containing LAB and yeasts are used for inoculation in most of the industrial processes (Guzel-Seydim et al., 2010; Mistry, 2004). This is due to difficulties in postfermentation requirements of the kefir grain separation at the end of fermentation. Using this method, activated starter culture is added to homogenized and pasteurized milk containing 2–5% milk fat. After fermentation at 25 °C for 20–24 h, the product can be stored at refrigeration temperatures up to 20 days (Guzel-Seydim et al., 2010).

2.5.2. Microbiota of kefir

Kefir grains are an excellent example of the co-occurrence of yeasts and bacteria (Mistry, 2004). The microbiota of kefir and kefir grains is affected from the ratio of kefir grains to milk, the origin and method of cultivation of the grains and the properties of substrates (Cetinkaya et al., 2010).
and Elal-Mus, 2012; Mistry, 2004; Witthuhn et al., 2005). Different types of milk as substrate affect the microbiota depending on their carbohydrate, fat, and protein content (Wszolek et al., 2001).

The microbiological properties of kefir samples were shown in Table 4. The Lactobacillus spp., Lactococcus spp. and yeast counts in kefir grains and kefir ranged from 1.0 × 10³ to 4.79 × 10⁶ cfu/mL, from 1.00 × 10⁵ to 1.000 × 10⁷ cfu/mL, and from <1.00 × 10⁵ to 7.94 × 10⁶ cfu/mL, respectively (Table 4). Garrote et al. (1998) reported that microorganisms released from the grain grew exponentially for 30 h in the case of yeasts and for 40 ± 5 h in the case of Lactobacillus spp., independent of the inoculum concentration. On the other hand, Lactococcus counts changed depending on the inoculum concentration and its count reached stationary phase after 20 h incubation.

The total percentage of LAB was found higher than the total percentage of yeasts by Witthuhn et al. (2005) and Magalhaes et al. (2011). In Brazilian kefir, the main group was LAB (60.5%) followed by yeasts (30.6%) and acetic acid bacteria (8.9%) (Magalhaes et al., 2011). The ratio of LAB to yeast in kefir grains was detected as 1000:1 (Guzel-Seydim et al., 2005). Witthuhn et al. (2005) reported that Lactobacillus spp. was the most frequently encountered microorganism in kefir grains. However, Lactococcus lactis subsp. lactis and S. thermophilus were determined as the predominating species in kefir grains (53–65%) and kefir samples (74–78%) by Simova et al. (2002). In Brazilian kefir, L. paracasei (35.7%) represented the largest and most commonly identified LAB, followed by L. parabuchneri (16.5%), L. casei (12.8%), L. kefir (12.4%) and Lactococcus lactis (9.6%) (Magalhaes et al., 2011). In addition to LAB, acetic acid bacteria such as Acetobacter lovaniensis (8.91%) (Magalhaes et al., 2011) and Acetobacter syzygii (Kesmen and Kacmaz, 2011) were isolated from kefir.

The microbial counts of traditional kefir and commercial kefir were different. The number of Lactobacillus spp. in traditional and commercial kefir was 7–8 log cfu/mL. Lactococcus spp., L. acidophillus and Bifidobacterium spp. counts in traditional kefir were 7 log cfu/mL, 7 log cfu/mL and 6 log cfu/mL, respectively. On the other hand, lower counts were obtained in commercial kefir samples, 5 log cfu/mL for Lactococcus spp., 3 log cfu/mL for L. acidophilus and 1 log cfu/mL for Bifidobacterium spp. (Bokzku et al., 2010).

Homofermentative and heterofermentative Lactobacillus spp., Lactococcus spp., Leuconostoc spp. and acetic bacteria and lactose-fermenting and lactose-nonfermenting yeasts were reported in kefir and kefir grains in other studies (Guzel-Seydim et al., 2010; Mistry, 2004). Among the yeast flora of kefir grains, non-lactose fermenting yeasts such as Candida spp. (Simova et al., 2002) and S. cerevisiae (Magalhaes et al., 2011; Simova et al., 2002) were dominantly isolated. The LAB and yeasts isolated from kefir and kefir grains are shown in Table 5. Lactobacillus kefiranofaciens and other lactobacilli in kefir grains produce a polysaccharide known as kefiran. Kefir grains consist of approximately 24% kefiran, which consists of glucose and galactose in equal proportion. The kefir beverage contains approximately 0.2–0.7% kefiran. It provides a slightly rosy texture to the final product (Mistry, 2004).

The antimicrobial activity of kefir and LAB isolated from kefir against a wide variety of gram-positive and gram-negative bacteria, and fungi was reported (Cevikbas et al., 1994; Garrote et al., 2000; Santos et al., 2003; Yukselгад et al., 2004). The antimicrobial activity of the kefir is related to lactic acid, volatile acids, hydrogen peroxide, carbon dioxide, diacetyl, acetaldehyde, and/or bacteriocins produced by LAB (Cetinkaya and Elal-Mus, 2012).

Probiotic properties of LAB isolated from kefir such as L. acidophillus, Lactobacillus helveticus, L. casei, Pediococcus dextrimucis, Pediococcus acidilactici, P. pentosaceus, Lactococcus cremoris, and Lactococcus lactis were evaluated by Sabir et al. (2010). It was determined that all Lactobacillus spp., Lactococcus spp. and Pediococcus spp. strains were able to survive at low pH values, at different bile salt concentrations, and were able to autoaggregate and coaggregate with E. coli. Santos et al. (2003) reported that L. acidophilus and L. kefiranofaciens had the best probiotic characteristics tested within the Lactobacillus spp. (L. kefir, L. brevis, L. paracasei, L. plantarum, L. acidophilus and L. kefiranofaciens).

High-quality probiotic kefir may be produced by the selection of starter cultures according to their antimicrobial and probiotic properties (Yukselгад et al., 2004).

2.5.3. Shelf life and safety of kefir

Kefir grains could become fully active after two or three propagations. It has a limited shelf life when left wet. During storage at 4 °C, kefir grains lose their activity within 8 to 10 days. However, dried grains are active for 12 to 18 months. Excessive washing and improper utilization alter the microbiota of grains and as well as the quality of the final product. For long time storage, kefir grains can be dried at room temperature for 36–48 h and stored in a cool and dry place or be kept in a frozen state (Mistry, 2004). Garrote et al. (1997) showed that the kefir produced from grains stored at −20 °C and −80 °C had the same microbiota and quality characteristics with kefir produced from unstored kefir grains. Freeze-dried kefir culture has been suggested for kefir manufacture to obtain uniform quality (Mistry, 2004).

Spoilage of kefir beverage could rapidly occur when contaminated grains with coliforms, Bacillus spp., Micrococcus spp. and mold were used in production (Mistry, 2004). Microbiological quality of 50 kefir samples was investigated and the average count of Lactobacillus, Lactococcus, Enterococcus, Enterobacteriaceae, S. aureus and yeast has been reported as 3.6 × 10⁵ cfu/mL, 1.8 × 10⁶ cfu/mL, 4.8 × 10⁴ cfu/mL, 7.3 × 10⁵ cfu/mL, 2.4 × 10³ cfu/mL and 7.7 × 10⁴ cfu/mL, respectively. Twenty four and 11 of 50 kefir samples were contaminated with coliform and E. coli, respectively (Cetinkaya and Elal-Mus, 2012). The contaminations of kefir samples with pathogenic bacteria such as E. coli and S. aureus possess health risks for consumers.

Fermentation can continue in kefir beverage during storage and cause extremely strong and undesirable products because of the relatively high residual lactose content and the presence of yeasts. Production of kefir with selected LAB and yeasts was performed to control the post-fermentation (Mistry, 2004).

The occurrences of biogenic amines in kefir samples were determined due to the activity of LAB. The occurrence of putrescine, cadaverine and spermidine was detected in all samples while tyramine was the prevailing biogenic amine. Total biogenic amine contents of kefir samples were between 2.4 and 35.2 mg/L. It was concluded that if consumption was higher than 0.5 L per meal, tyramine might cause a mild adverse event for consumers treated with monoamine oxidase inhibiting drugs (Ozdestan and Üren, 2010b).
The rheological behavior of a food matrix is an important physical property that has a direct association with product overall quality and processing characteristics, as well as on consumer acceptability (Yegin and Fernandez-Lahore, 2012). In addition, determining any given component functionality in product development, shelf-life of the food and evaluating the food texture by correlate to sensory properties require rheological data (Steffe, 1996). In summary, rheology is related to food acceptability, food processing and handling (Bourne, 1982).

Foods are generally classified as Newtonian or non-Newtonian depending on the relationship between shear stress and shear rate. The Newtonian behavior is expressed by the Newton’s law of viscosity whereas one of the non-Newtonian behaviors, pseudoplasticity (shear thinning), can be modeled using a power-law equation. The following equations are used for Newtonian and pseudoplastic fluids, respectively:

\[ \tau = \eta \gamma \tag{1} \]

\[ \tau = K\gamma^n \tag{2} \]

where \( \tau \) is shear stress (Pa), \( \eta \) is dynamic (apparent or absolute) viscosity (Pa s), \( \gamma \) is shear rate (s\(^{-1}\)), \( K \) is consistency index (Pa s\(^n\)), and \( n \) is flow behavior index (-). The viscosity of a non-Newtonian material must be presented with the shear rate at which the viscosity is recorded along with the temperature. However, it has been observed that neither shear rate nor temperature was not reported for viscosity results for the beverages. In addition, for some of the beverages such as hardaliye and shalgam juice there is no rheological study whereas the rheological studies about ayran, kefir and boza are relatively limited.

It was reported that boza exhibited pseudoplastic behavior and there was a correlation between viscometric constant and dry matter content of boza. It was suggested that the consistency coefficient of boza can be used as a predictor of sensory scores of mouthfeel and appearance. Furthermore, it was predicted that rheological parameters...
of boza may be correlated with pH, acidity and flavor (Genc et al., 2002). Genc et al. (2002) studied the rheological parameters of boza commercially obtained from local markets in Istanbul, Ankara and Izmir. They reported that K and n values of boza samples at 10 °C in between 3.125–21.467 Pa s⁻¹ and 0.267–0.502, respectively. They also prepared boza with various dry matters under laboratory conditions using boza obtained from a market for inoculation. They gave the K and n values for laboratory samples as in between 0.830–11.617 (Pa s⁻¹) and 0.3053–0.4481, respectively. These results indicated that consistencies of commercially obtained boza are higher than boza prepared in laboratory. In the same study, panelists had higher scores for boza with high consistency index as a general trend (Genc et al., 2002).

The rheological studies showed that ayran exhibits pseudoplastic (shear thinning) or thixotropic behavior depending on its fat content (Köksoy and Kilic, 2003; Lokumcu et al., 2002). In the study, the whole ayran had pseudoplastic behavior whereas light ayran that does not contain fat exhibited nearly Newtonian behavior (Bayraktaroglu and Obuz, 2008). In another study, it was reported that it is unknown whether any potential differences in the physical, chemical and microbiological properties of ayran samples arose from the yoghurt properties produced by different methods (Kocak et al., 2006). Lokumcu et al. (2002) studied rheological properties of ayran obtained from local markets in Istanbul. They reported that K and n values of ayran samples at 9.5 °C depend on their fat contents in between 0.07–0.70 Pa s⁻¹ and 0.38–0.66, respectively. They concluded that rheological properties of ayran samples sold in Istanbul varied due to different compositions of products and/or manufacturing technologies. Köksoy and Kilic (2003) prepared ayran in the laboratory by the traditional method of mixing yoghurt with water and salt. They found that K values of ayran samples decreased from 1.214 Pa s⁻¹ to 0.018 Pa s⁻¹ with the addition of salt up to 1% and water up to 50% at 10 °C. In contrast, n values increased from 0.297 to 1.004 at 10 °C with the addition of the same amount of salt and water (Köksoy and Kilic, 2003). These results showed how the components affect the rheological properties of ayran. Janhoj et al. (2008) studied rheological characterization of acidified milk drinks (AMD) at 12 °C. They specified AMD which did not contain salt, as a special variety of the Turkish product, ayran. They reported that K and n values of AMD acidified with LAB as 0.0437 (the unit of K was not given) and 0.6161, respectively. This sample had 2% non-fat milk solids and 0.5% pectin. The K and n values of sample with 2% non-fat milk solids and 0.5% carboxymethyl cellulose were 0.0280 (the unit of K were not contain fat exhibited nearly Newtonian behavior (Bayraktaroglu and Obuz, 2008). Genc et al. (2002) studied the rheological parameters of boza prepared in laboratory. In the same study, panelists had higher scores for boza with high consistency index as a general trend (Genc et al., 2002).

3.1. Fermentation–structure relations

An understanding of structure–function and/or property relations of individual components in a mixed system containing protein and polysaccharide is of particular interest for creating use of functional ingredients in foods (Sharma et al., 2011). In this context, this approach can be extended to the understanding of the structural characteristics of the fermented foods. For instance, both ayran and yoghurt are made of milk which is a Newtonian fluid. The same starter cultures are used for their lactic acid fermentation. During yoghurt fermentation, milk as a liquid turns into yoghurt which is a three-dimensional viscoelastic gel; somewhere in between ayran can be obtained as a pseudoplastic or thixotropic fluid depending on its fat content. Pseudoplasticity, thixtropy and viscoelasticity are very different rheological behaviors; their flow characteristics are different as well as their perceptions in the mouth. If the structure–function relations for fermentation process would be constituted then it can be determined that at which point and under what conditions these transitions take place.

Most of the studies related to the traditional fermented foods have been focused on the fermentation process in terms of microbiota. On the other hand, the studies on the quality characteristics of fermented foods have been devoted to the determination of composition, color characteristics, rheological parameters and sensory properties. In these studies, researchers have suggested composition-related solutions to the quality-related problems of fermented beverages. For instance, Dogan (2011) suggested that maintaining the texture can be a problem in commercial manufacture of fermented dairy products and keeping the quality of the final product increasing milk solids may solve the textural problem. In another case, weak aroma and taste, low viscosity, sedimentation and serum separation are the main problems especially in semi-skimmed and skimmed ayran. The general trends in industrial applications and scientific studies for solving these problems are included in the addition of stabilizers and gums such as whey, locust bean gum, high methoxyl pectin, gelatin and guar gum (Bayraktaroglu and Obuz, 2008; Köksoy and Kilic, 2004). In addition, utilization of transglutaminase for crosslinking of proteins which are considered as the responsible molecules for the texture in ayran was reported for obtaining samples with higher viscosity and lower serum separation (Sanli et al., 2011). However, it appears that the effects of fermentation conditions such as substrate, time, temperature, microbiota and duration on the structure of fermented products were overlooked. This is mainly due to the finishing of the fermentation which is applied based on the general sensory properties and/or acidity of the fermented foods. Actually, fermentation process is responsible for the structure of the product, which is neglected in most cases. Therefore, as a starting point, the exopolysaccharide (EPS) studies of starter cultures can be taken into account. In the following part the studies are reviewed for effects of fermentation process on quality characteristics of fermented beverages and properties of EPSs produced by LAB.

3.2. Effects of fermentation conditions

The problems in boza are related to manufacturing process which is non-standard and with low technology and lacking in modern equipment. Boza that is inoculated with old boza is preferred over boza that is obtained using pure starter culture. The cold storage and the additives affect the quality of boza (Yücel and Ötes, 1998).

Serum separation and viscosity are important quality characteristics in ayran and similar fermented milk products (Sanli et al., 2011). In addition, all five beverages may deteriorate during storage due to acidity development, and serum separation and/or solid sedimentation problems compared to kefir with skim milk powder. They explained that kefir with whole milk powder had lower G″ and G′ due to its high fat content compared to the sample with skim milk powder.
which can be related to action of microorganisms and their fermentation conditions.

Microbial distribution of kefir grains, grain–milk ratio, incubation time and temperature, sanitation during separation of kefir grains, washing of kefir grains and cold storage drastically affect the product quality and the microbiota of the kefir grains (Guzel-Seydim et al., 2005). It was reported that it is usually thought that kefir grains from different countries/origin have different microbiota; however, the LAB and yeast contents of kefir grains are similar (Kök-Tas et al., 2012).

Recently, the characterization studies of exopolysaccharides (EPS) produced by microorganisms to develop texture of various foods have increased. The reason for choosing the EPS from LAB over plant polysaccharides used as thickening and stabilizing agents is that polysaccharides from plant sources are not acceptable in dairy products if they have an “all dairy” label. EPS from LAB have been reported to be used as stabilizing, viscosity modifying and gelling agents in foods (Ahmed et al., 2013). In similar studies the authors suggested that the addition of EPS from LAB to various foods for modifying rheological properties by isolation of bacteria from fermented food and then the addition of EPS after purification. However, it seems that no studies have conducted about how fermentation process during manufacturing of fermented foods affects the rheology of the product in terms of production of EPS. According to the current literature of the characteristics of EPS they can be manipulated by using fermentation parameters such as substrates, temperature, time and pH. Examples of such studies are summarized in Table 6.

4. Conclusions

The variations in qualities of traditional fermented beverages were affected from several factors such as utilization of different raw materials, manufacturing methods, natural microbiota and fermentation conditions. The problems in these beverages such as serum separation/solid particle sedimentation, acidity development and low viscosity affect consumer acceptance in terms of mouthfeel and appearance, and are related to quality characteristics during manufacturing, transportation and storage. It appears that most of the quality problems are rheology-related which are affected by initial raw materials and composition of product, fermentation conditions, type of microorganisms and their numbers and ratios. The fermentation ability and fermentation products such as EPS vary depending on microorganism species, leading fermented products with different rheology. By using LAB and yeast isolates as starter cultures, controlled fermentation studies can be carried out to optimize quality for obtaining stable products. The selection of strains with probiotic and antimicrobial properties also enhance the functional properties of traditional foods. Apparently, more studies are needed to investigate the fermentation, rheology and their relations for improving quality and safety of fermented beverages.

As a conclusion, in order to maintain standardized and efficient production future studies about traditional fermented non-alcoholic beverages should focus on:

- Selection of appropriate starter cultures and determination of their optimum growth conditions,
- Selection of appropriate microbiota, ratios and numbers of microorganisms in the microbiota,
- Selection of microbiota for functional fermented beverages with desired probiotic and antimicrobial properties and minimum biogenic amine formation,
- Effects of fermentation process parameters, i.e., substrate, time, temperature, EPS production on quality defects such as sensory problems, serum separation, solid sedimentation and low viscosity using rheology as a tool, which may be used for new product developments such as more creamy or spreadable kefir.

### Table 6

Exopolysaccharides obtained from LAB under various substrate and fermentation conditions, and their characteristics.

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Substrate</th>
<th>Fermentation conditions</th>
<th>Isolated EPS</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactobacillus helveticus BCRC14030</td>
<td>Reconstituted skim milk (10% w/v)</td>
<td>0–84, 37°C, pH 5.0</td>
<td>Highest EPS yielded as 0.73 g/l from L. helveticus BCRC14030 at 60 h</td>
<td>Lin and Chien (2007)</td>
</tr>
<tr>
<td>L. helveticus BCRC14076</td>
<td></td>
<td></td>
<td>EPS had 5.5 × 10^10 Da molecular weight, 14.2% solubility, 496% water holding capacity, 884.5% oil binding capacity and 93.4 °C melting point</td>
<td>Ahmed et al. (2013)</td>
</tr>
<tr>
<td>Streptococcus thermophilus BCRC14085</td>
<td>Liquid whey</td>
<td>72, 30°C</td>
<td>Highest yield of kefiran polymer was obtained at conditions of 24 h, 25 °C and 80 rpm agitation rate with the presence of all type of enrichments.</td>
<td>Zajsek et al. (2013)</td>
</tr>
<tr>
<td>L. kefirans LZW3</td>
<td></td>
<td></td>
<td>After fermentation, contents and viscosities of soluble fibre in oat fibre concentrates decreased while molecular weight was not affected.</td>
<td>Lambo et al. (2005)</td>
</tr>
<tr>
<td>Commercially obtained kefir grain</td>
<td>UHT full fat cow milk enriched with carbohydrates/nitrogen sources/minerals</td>
<td>24, 25°C or 43°C</td>
<td>Highest yield of kefiran was obtained at conditions of 24 h, 25 °C and 80 rpm agitation rate with the presence of all type of enrichments.</td>
<td>Ahmed et al. (2013)</td>
</tr>
<tr>
<td>Pediococcus damnosus 2.6, L delbrueckii subsp. bulgaricus, Streptococcus salivarius subsp. thermophilus, L. acidophilus</td>
<td>Native oat and barley fibers with glucose or sucrose addition</td>
<td>20, 28°C or 37°C</td>
<td>After fermentation, contents and viscosities of soluble fibre in oat fibre concentrates decreased while molecular weight was not affected.</td>
<td>Ahmed et al. (2013)</td>
</tr>
<tr>
<td>Kefir grains</td>
<td>Cheese whey supplemented with yeast extract</td>
<td>120, 15–35°C, pH 3.5–7.5</td>
<td>Kefiran production was optimized at lactose concentration of 67 g/l, yeast extract of 13 g/l, pH 5.7 and 24 °C. EPS exhibited antioxidant activity</td>
<td>Ghasemlou et al. (2012)</td>
</tr>
<tr>
<td>Lactococcus lactis subsp. lactis</td>
<td>MRS medium</td>
<td>24, 37°C</td>
<td>The optimum temperature for EPS production was 30 °C</td>
<td>Guo et al. (2013)</td>
</tr>
<tr>
<td>Commercially obtained kefir grains</td>
<td>Pasteurized skim milk with addition of ammonium citrate, calcium chloride and lactose monohydrate</td>
<td>120, 35–45°C, pH 5.0</td>
<td>Free EPS increased the stiffness of acid milk gels.</td>
<td>Mendes et al. (2013)</td>
</tr>
<tr>
<td>Streptococcus thermophilus ST143</td>
<td>L. plantarum CBS</td>
<td>24, 40°C, 6.0</td>
<td>Free EPS increased the stiffness of acid milk gels.</td>
<td>Mendes et al. (2013)</td>
</tr>
<tr>
<td>Bifidobacterium longum subsp. infantis CCUG 52486 and Bifidobacterium infantis NCIMB 702205</td>
<td>Sterile reconstituted skim milk supplemented with casein hydrolysate MRS agar</td>
<td>18, 37°C</td>
<td>EPS added yoghurt showed lower syneresis and high storage modulus and firmness</td>
<td>Prasanna et al. (2013)</td>
</tr>
<tr>
<td>L. plantarum CBS</td>
<td></td>
<td></td>
<td>EPS composed of galactose and glucose and exhibited significant antioxidant activity.</td>
<td>Zhang et al. (2013)</td>
</tr>
</tbody>
</table>
-- Application of Hazard Analysis & Critical Control Points (HACCP) system for food safety.

-- Mycotoxin contamination of traditional fermented non-alcoholic beverages, which are limited in the literature.

References