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# Fermented Traditional Probiotic Beverages of Turkish Origin: A Concise Review

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#### ABSTRACT

The fact that foods can provide additional medicinal health benefits beyond basic energy requirements and nutritional needs for survival, has been proven through rigorous scientific researches over the years. Although Turkey is historically believed to be the ancient origin of a decent number of fermented probiotic foods and beverages, there is still little awareness amongst the overall populace regarding the usefulness of these medicinal products also known as functional foods. Hence, the current review article discusses probiotics as a top notch variety of functional foods and encapsulates their history as well as some proven beneficial effects of some known probiotic microorganisms on human health. More importantly, greater emphasis is placed on elucidating scientific data on seven fermented traditional probiotic beverages of Turkish origin namely boza, kefir, ayran, shalgam, hardaliye, koumiss and gilaburu juice. The paper concisely describes their main characteristics, probiotic microbiota composition, production techniques as well as nutritional properties and some potential health benefits derivable from their consumption.

## **Probiotics concept: An overview**

The term "probiotic" has its roots in the Greek word "probios" meaning "For Life". After knowledge of the existence of probiotics came to the forefront, several definitions have been proposed by various experts and scientists [1, 2, 3].

The most widely accepted definition given by the World Health Organization (WHO) and currently in use today describes probiotics as "Live microorganisms which, when administered in adequate amounts, confer a health benefit on the host." [4].

#### **Prebiotics and synbiotics**

Prebiotics and synbiotics are the two terms that usually appear to play a complementary role wherever probiotics are mentioned or used. Prebiotics are non-digestible food components

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which improve the health condition of a host via stimulation of the activity and/or growth of a limited number of microorganisms resident in the gastrointestinal tract (GIT) [5]. Prebiotics are also described by experts as non-viable food constituents that impart a benefit to the host's health through the modulation of gut microbiota [6]. Although prebiotics are found in natural products, they may also be added to foods in order to improve their health and nutritional values. Several foods viz. asparagus, banana, chicory roots, onions, garlic, tomato, alliums and Jerusalem artichoke are very rich in the prebiotics fructo-oligosaccharides (FOS) and lactulose whereas raffinose and stachyose are naturally present in beans and peas [7].

The term "Synbiotic" refers to a combination of probiotics and prebiotics acting synergistically to improve the host's health [5]. They are also defined further as mixtures of prebiotics and probiotics which beneficially impart the host's health by improving the implantation and survival of live microbial dietary supplements in the host's GIT [8].

Typical examples of synbiotic combinations used in nutrition include: *Lactobacillus* + inulin; *Bifidobacterium* and *Lactobacillus* + inulin; *Bifidobacterium*, *Lactobacillus* and *Streptococcus* + FOS; *Bifidobacterium* and *Lactobacillus* + oligofructose [9].

## History of probiotics at a glance

The knowledge of the benefits of lactic acid fermentation on man's health dates back to prehistoric times. The Bible has records of sour milk consumption by our forefathers, and in fact, it is believed by Persian traditions that Abraham owed his longevity and fecundity to habitual yogurt consumption [70]. The Bible in Genesis 18:8 also reveals further that Abraham did not only consume fermented sour milk but also customarily entertained guests with it. Furthermore, several records have shown that between 2000 and 3000 BC, fermented milk products (milk, butter and cheese) were commonly used in different civil establishments such as Egyptians, Indians, Greeks and Romans [10]. In ancient Greece and Rome, fermented milk consumption was recommended for children and convalescents. The Roman naturalist Pliny the Elder also suggested the consumption of fermented milk for the treatment of intestinal problems [10].

Even though the term "probiotics" which is currently being used today to describe bacteria associated with beneficial health effects, was not coined until mid-1960, the unprecedented works done by Henry Tissier and Eli Metchnikoff in the beginning of the twentieth century

had already given some inspiring shreds of scientific evidence towards the probiotics concept. A French pediatrician at the Pasteur Institute in Paris, Henry Tissier in 1906 reported that infants with abundant bifidobacteria in their intestinal system had minimal gastrointestinal troubles, as evidenced by low diarrheal problems. On the contrary, stool samples of children suffering from diarrhea were found to have a very low amount of these "bifidobacteria" [11]. Tissier, therefore, suggested the administration of these bifidobacteria to patients suffering from diarrhea in order to help restore a healthy gut microflora. In the same vein, in 1908, Eli Metchnikoff, a Russian immunologist and scientist also at the Pasteur Institute in Paris, won the Nobel Prize for medicine for his research revealing that destructive microbes could be substituted by beneficial ones to treat intestinal disorders and illnesses. Metchnikoff had observed that the countryside residents of Bulgaria lived long in spite of severe poverty and unfavourable climatic conditions. They had an average lifespan more than the richer Europeans, and he reported that they consumed fermented milk products heavily populated with lactic acid bacteria (LAB). He hypothesized that those LAB offered a special kind of protection against intestinal autointoxication. Metchnikoff, therefore, concluded that the LAB present in the fermented milk products had antiaging health benefits [12]. The pioneering works of these two scientists - Henry Tissier and Eli Metchnikoff are known to be the studies that established scientific hypotheses regarding the existence and use of probiotic microbes.

#### History of probiotics at a glance with the Turkish and Turkic People

The ancient Turkish people who dwelt as nomads in Asia were the first to make yogurt which was called "yoghurut" (Turkish: yoğurt) [64]. Hence, the name "yogurt" as popularly known today is widely documented to have originated from the Turkish word "yoğurmak", meaning "to coagulate, thicken, or curdle" [65]. The Turkish-originated *yogurt* which had been invented in modern day eastern Anatolia (Eastern Turkey) - a part of the erstwhile Mesopotamia - dates back to five thousand years ago. Furthermore, the use of yogurt by medieval Turks was also recorded in the books Diwan Lughat al-Turk and Kutadgu Bilig which were both written in the 11th century [66, 67, 68]. The word "yogurt" was mentioned in the texts and its use by nomadic Turks was reported. The Turks were also reported as the

first to evaluate the medicinal use of yogurt for various illnesses and disease symptoms, such as diarrhea and cramps, and for alleviation of the discomfort from sunburnt skin [68].

## Probiotics as an ideal and typical functional food

The term "Functional Food" refers to special foods that have curing and/or preventive effects beyond their nutritional value. In other words, functional foods are substances or supplements which are consumed to obtain a specific result [13].

There are varieties of functional foods recently developed many of which are being produced all around the world. These include foods enriched with prebiotics, probiotics, synbiotics, anthocyanins, phytosterols, antioxidants, and isoflavones as well as other functional foods like salt-reduced, fat-reduced, and sugar-reduced foods. The first choice to exert beneficial effects on human health among these foods are functional probiotic foods [14].

Table 1 highlights the proven beneficial health effects of some globally studied probiotic strains.

Probiotic Group	Probiotic Species	Proven Benefits on Human Health		
	Lactobacillus casei	. Treatment of lactose intolerance;		
	Shirota	. Treatment of antibiotic, bacteria and virus-associated diarrhea		
Lactobacilli (i.e. rota		(i.e. rotavirus, Clostridium difficile).		
	Lactobacillus	. Remediation of inflammatory conditions via the modulation of		
	rhamnosus	GIT microbiota;		
	GG	. Reduction of the duration of antibiotic- (erythromycin) induced		
		diarrhea;		
		. Downregulation of the immune-inflammatory response in milk		
		hypertensive subjects.		
	Lactobacillus casei	. Stimulation of the immunity of the GIT;		
		. Alleviates the symptoms of Crohn's disease;		
		. Reduction of the severity and duration of diarrhea;		
		. Exhibition of strong antimicrobial activities.		
Bifidobacte	Bifidobacterium	. Inhibition of enteropathogen-cell interactions by adhering to		
ria	breve	human intestinal epithelial cells;		
		. Augmentation of anti-rotavirus or anti-influenza virus IgA		
		production;		
		. Activation of the humoral immune system.		

<i>bifidum</i> . Prevention of infectious diarrhea ( which is observed as reduced shedd . Competing successfully for nutrien putrefactive or pathogenic bacteria; . Increases seroconversion rates and . Secretion of acetic and lactic acids		<ul> <li>Reduction of the incidence of diarrhea;</li> <li>Prevention of infectious diarrhea (by a small protective effect which is observed as reduced shedding of rotavirus);</li> <li>Competing successfully for nutrients and space against putrefactive or pathogenic bacteria;</li> <li>Increases seroconversion rates and antibody responses;</li> <li>Secretion of acetic and lactic acids (which help the inhibition of invasive pathogens like <i>Candida</i> spp. and <i>Escherichia coli</i>).</li> </ul>
	Bifidobacterium animalis	<ul><li>Reduction of the risk of acute diarrhea in human;</li><li>Normalization of the intestinal motility of obstipated subjects.</li></ul>
Yeasts	Saccharomyces cerevisiae Boulardii	<ul> <li>Reduction of the risk and the duration of antibiotic associated diarrhea (AAD);</li> <li>Prevention of traveler's diarrhea. It alters the GIT microbiota in a healthy mode and treats AAD or <i>Clostridium difficile</i> diarrhea;</li> <li>Inhibition of the development of pathogenic colitis and enterocolitis.</li> </ul>

\*Adopted and modified from [15]

# Fermented traditional probiotic beverages of Turkish origin

Up to date, there are only a few individual reports on each fermented traditional Turkish beverage encountered in the literature [16, 17, 18, 19, 20] and two reviews highlighting some popular products [21, 25]. Thus, the current paper aims to fill in the existing knowledge gap by combining reports of individual studies in a clear, concise and unprecedented manner, and holistically reviewing all the major fermented traditional probiotic drinks (kefir, ayran, boza, hardaliye, shalgam, gilaburu juice, and koumiss) produced and consumed in different regions of Turkey. The study provides concise information with regards to their characteristics, production techniques, probiotic microbiota composition, nutritional data and health benefits.

# Kefir

Kefir is a slightly acidic mildly alcoholic fermented beverage which is widely consumed in Turkey, and it has been described as the 21<sup>st</sup> century yoghurt [22]. The name "kefir" originates from the Turkish word "Keyif" which means "Joy/Pleasure" and it is used to describe the contentment experienced from drinking this beverage [23, 24]. Kefir can be produced by the fermentation of milk from cow, goat, ewe, or other sources [25]. For example, milk derived from plants such as coconut has been used to produce kefir in India and some other parts of the world [26]. Traditionally, it is produced by inoculating the starter culture "kefir grains" (i.e. a consortium of bacteria and yeasts in a polysaccharide matrix) to

pasteurized milk [24, 27]. The microbial constituents of kefir grains include lactic acid bacteria (LAB), lactose-fermenting yeasts (produce ethanol and CO<sub>2</sub>), acetic acid bacteria (AAB), and non-lactose fermenting yeasts. The LAB are mainly responsible for lactose conversion into lactic acid, resulting in a pH drop and enhancing preservation [28]. Kefir's main fermentation products include ethanol, lactic acid and CO<sub>2</sub>, which confer on the beverage low alcohol content, acidity and viscosity. Some minor components such as amino acids, acetaldehyde, diacetyl and ethyl may also be produced, contributing to its flavor composition [28]. The microbiological quality and physicochemical properties of kefir are influenced by the microbial property of used kefir grains, the grains to milk ratio, incubation temperature and time, storage temperature and sanitation conditions [29]. Kefir has been reported to exhibit antimicrobial activity which is attributable to lactic acid, bacteriocins, hydrogen peroxide, acetaldehyde, volatile acids, diacetyl, and/or carbon dioxide produced by the bifidobacteria and the LAB present in the drink [30].

The traditional procedure for kefir production is described in figure 1 below.

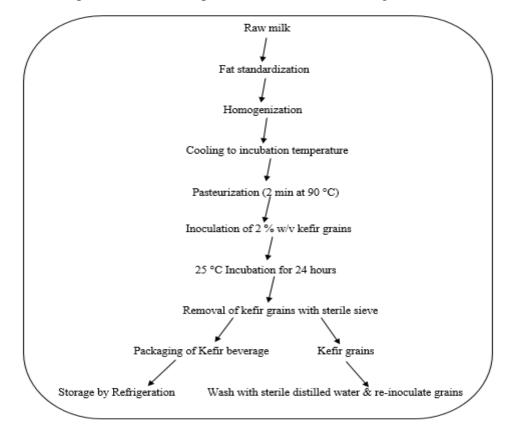


Fig 1 Traditional method of kefir production [29]

#### Ayran

Ayran otherwise known as yoghurt drink is a delicious fermented milk beverage. It is produced by either the addition of water to yoghurt or by inoculation of starter cultures containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus* to pasteurized and standardized milk [31]. In other words, the blending of yoghurt with 30–50 % of water and 0.5–1 % of salt results in the generation of the beverage called ayran [32]. Ayran is consumed widely as a national drink across the length and breadth of Turkey. It is a highly valuable and readily digestible drink that is rich in calcium and vitamin. The chemical composition of ayran depends on the kind of milk used, the efficiency of fat removal and the dilution rate [25].

It has been proven that by using inulin as prebiotic and *L. acidophilus* and *Bifidobacterium* spp as the probiotic strains, the improvement of ayran's functional and probiotic properties is possible. This improved product was reported to have a better taste and higher *Lactobacillus* count [33].

The two different methods of ayran production are highlighted in figure 2 as follows.

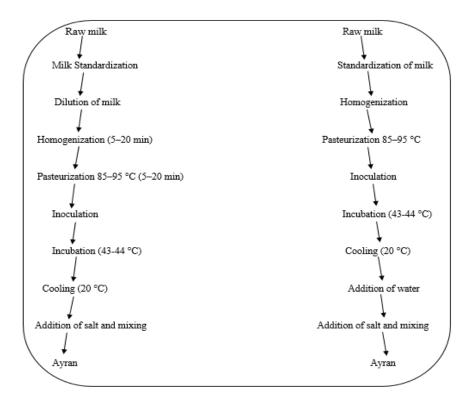


Fig 2 Two industrial techniques for ayran production [25, 34]

#### Boza

Boza is a fermented traditional non-alcoholic Turkish beverage manufactured by a combination of yeast and lactic acid fermentation of cereals or grains such as maize, millet, rice semolina, rye or wheat, and mixed with sugar or saccharine. It is consumed widely in Turkey and many Balkan countries owing to its pleasant flavor, taste and nutritional properties [35].

The two kinds of fermentation that occur in boza production are lactic acid fermentation by LAB as well as alcoholic fermentation by yeasts. Antimicrobial metabolites such as lactic acids are produced by the LAB and these increase acidity which imparts a preservative effect whereas the metabolites produced by the alcoholic fermentation of the yeasts bring about the odor and mouthfeel of boza [25, 37, 38]. The utilization of different kinds and amounts of cereals and/or cereal products as raw materials as well as the uncontrolled fermentation conditions can result in different compositional variations in boza drinks [35, 36]. The production technique of boza is briefly described in figure 3 below.

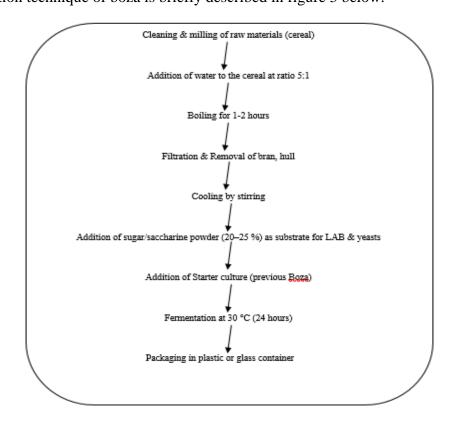


Fig 3 Industrial production of boza [37, 38]

## Hardaliye

Hardaliye is a red grape-based fermented traditional non-alcoholic beverage produced in the Thrace region (the European side of Turkey) [39]. Hardaliye has been classified as a non-dairy probiotic beverage due to its rich LAB flora [40].

The nutritional value of hardaliye emerges from grapes and the fermentation process. The functional properties and health benefits of hardaliye are attributable to its ingredients i.e. grapes, mustard seeds (contain etheric oils, allyl isothiocyanate and sinigrin which is a carcinogenesis-suppressing agent). Grapes are known to be very rich in phenolic contents, thus hardaliye provides high antioxidant effects, thereby preventing oxidative stress and contributing to the inhibition of carcinogenesis. Furthermore, hardaliye's high phenolic content and strong antioxidant capacity also decreases serum homocysteine concentrations [41]. Besides its nutritional value, hardaliye also aids digestion system and helps in the prevention of coronary heart disease [40]. In addition, mustard oils have been found to exert medicinal effects on circulatory disorders, common cold and bronchitis, and possess antimicrobial properties [41]. One glass of hardaliye contains 39 g of carbohydrates and supplies 170 kcal of energy. Owing to its palatable, salt-free, nonalcoholic, low-fat and non-dairy characteristics, it is suitable for consumption by vegetarians, children, hypertensive, dairy intolerant, and people with high cholesterol levels [41].

The traditional technique of producing hardaliye is described in figure 4 below.

## Shalgam (Şalgam)

Shalgam is a cloudy, red-colored and sour black carrot juice that is produced by lactic acid fermentation of a mixture of black carrot, bulgur (broken wheat) flour, turnips, salt and water. The raw materials of shalgam juice are rich in sugars that can be fermented by yeasts and LAB. Shalgam juice is a highly nutritional beverage due to its high mineral, amino acid, polyphenol, and vitamin contents [44, 45]. It is reported to possess 88.3–114.1 mg/L of the anthocyanin called cyanidin-3-glycoside. The extraction of this compound from black carrot into the shalgam juice occurs during processing [46].

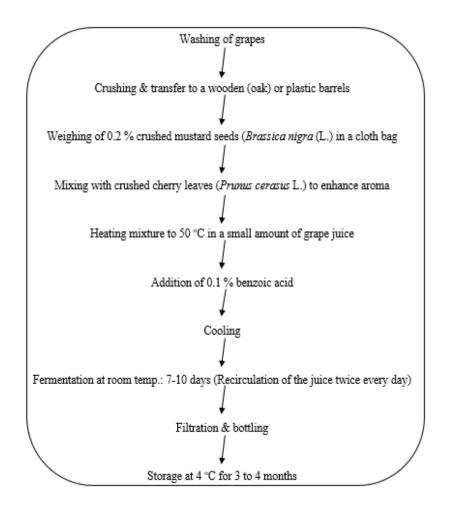


Fig 4 Industrial production of hardaliye [42, 43]

According to Yilmaz-Ersan and Turan's study, the mineral contents of shalgam juices obtained from domestic markets in Bursa, Turkey comprise calcium, phosphorus, magnesium, potassium, and sodium as the major elements, with about 1 mg/L of heavy metals [47]. The traditional technique of shalgam production is briefly explained in figure 5 below.

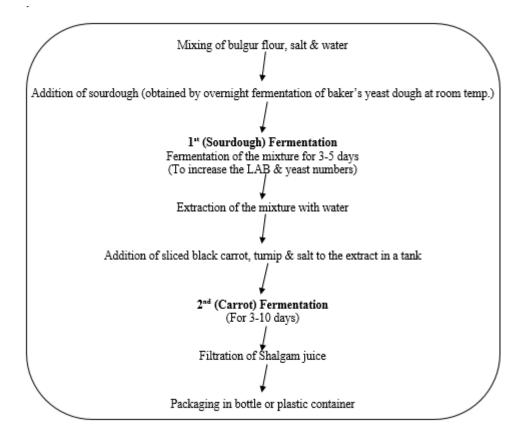


Fig 5 Traditional method of shalgam production [45, 48, 49]

## Gilaburu juice

Gilaburu juice is a fermented fruit juice made from the European cranberry bush (*Viburnum opulus* L.) which is the fruit of a deciduous shrub belonging to the family Caprifoliaceae. It is mainly grown indigenously in the Central Anatolia Region of Turkey, and traditionally the fruits are collected at the end of autumn. The barks and fruits of European cranberry bush tree have wide use in pharmacology. Its antispasmodic properties were independently discovered by European, Asian and Native American people. In addition, it is also being used for relief from asthma, fever, cold, menstrual cramps, stomach ache, nervousness, cough, blood pressure, water retention problems, infertility and uterine infections [50].

Fermented gilaburu (European cranberry bush) juice may be considered as a potential functional food as it contains a couple of antioxidants such as anthocyanins, ascorbic acid, flavonoids and phenolic acids at high levels. Furthermore, considering the large amounts of

LAB species present in the fermented fruit juice, it may also be regarded as a potential probiotic product [16]. Owing to its astringent taste, this juice has low acceptability but it is however believed that the local consumers can suppress its extremely sour and bitter taste by a longer fermentation period [51].

The traditional method of gilaburu juice production is summarized in figure 6 as shown below.

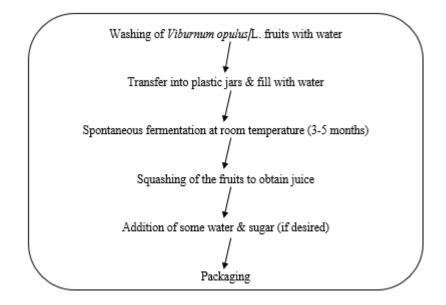


Fig 6 Traditional method of gilaburu juice production [51]

#### Koumiss

Koumiss also known as "Kımız" in Turkish is a mildly alcoholic and sour traditional dairy product which is produced from the fermentation of mare's milk. Koumiss which is believed to have first originated from the Central Asian nomads possesses a smell similar to that of sour buttermilk or sour cheese and gives foamy appearance when shaken. The name "koumiss" is also mentioned in Divan-Lugat at-Turk where it is described by Kashgari Mahmut as "soured mare's milk" and it is found in Turkish dialects till today [66]. Even though it is a significant beverage especially for Turkish, Bulgar and Mongol peoples, it is also valuable for the people of Kazakhs, Kyrgyz, Uygurs and Yakuts origin [60]. It is especially widely consumed in Mongolia as a national drink and in Russia for its therapeutic values. Slightly similar to kefir but in contrast to the solid kefir starter, i.e. "kefir grains", koumiss is made from a liquid starter and has a sharp acidic and alcoholic taste [69]. Owing to the limited availability of mare's milk, cow milk which is richer in fat and protein content but lower in lactose than mare's milk, is now being used for the commercial manufacture of koumiss [52]. Koumiss is a long-lasting drink, thus it has been consumed for many years by nomadic Turks both for quenching thirst and for health improvement [69].

The production technique of koumiss is described in figure 7 as follows.

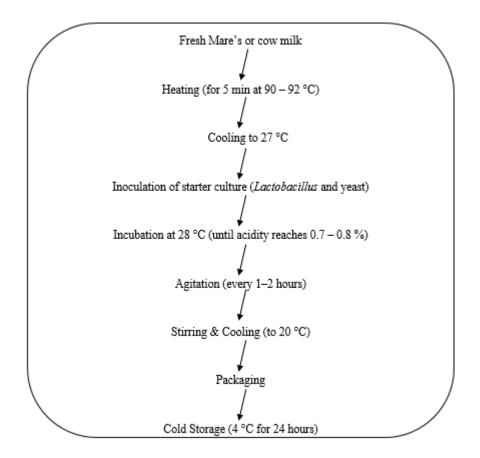


Fig 7 Steps involved in the production of koumiss [52]

Lastly, table 2 summarizes the available scientific data on the seven major fermented traditional probiotic beverages of Turkish origin giving concise information about their sensory or organoleptic properties, raw materials, nutritional data and probiotic microbiota composition.

Beverage	Nutrition Data	Sensory Property and	Substrate	Microbiota/Probiotics
		Nature		
Kefir	Per 100 g: fat $1.5$ g, protein $3.0-3.4$ g, and lactose $2.0-3.5$ g, lactic acid $0.6-1.0$ ml/100 ml(19), vitamins B1, B12, B6, K, ascorbic, folic acid, amino acids, Mg, Ca, P	Effervescent milk, Sour, acidic and slightly alcoholic, viscous, self-carbonated, elastic consistency, and uniform creamy, perceivable yeast aroma, tart flavour, slightly foamy body with white/yellowish colour	All kinds of milk (cow, goat, camel, sheep, buffalo) and milk substitutes, e.g. soya milk, coconut and rice milk	Lactobacillus. brevis, L. aci bulgaricus, L. plantarum, L. cas helveticus, L. buchneri, thermophilus, S. durans, Leuc Lactococcus lactis, L. cremoris, Pediococcus acidilactici, Kluyv lactis, Candida inconspicua, C Cryptococcus humicolus
Ayran	Total DM ( $1.07-11$ %), fat ( $0.1-3$ %), protein ( $1.44-3.48$ %), salt ( $0.17-1.75$ %) and high content of vitamins and Ca	Low viscosity and non- alcoholic	Cow Milk or others	Streptococuus thermophilus, La subsp. Bulgaricus
Boza	Lactic acid, carbohydrate, protein, vitamins and fibre such as pyridoxine, riboflavin, thiamine, and nicotinamide	Colloid suspension, Viscous liquid, slightly sweet or sour flavour, pale yellow, acidic and alcoholic odour	Millet flour, maize, rye, oat, and barley flour or Wheat and rice semolina	Lactobacillus plantarum, L. bro fermentum, L. coprophilus, Lacto mesenteroides, Lc. reffinolactis, cerevisiae, Candida glabrata Geotrichum candidum and Geotri
Hardaliye	Antioxidants, phenolic content, Sinigrin (from mustard seeds)	Acidic, non-alcoholic, original colour of the grapes	Red grape juice and Pomace	Lactobacillus paracasei, L. brev casei ssp. Pseudoplantarum; L. po
Shalgam	Vitamins (A, C, and B), minerals (Ca, K, and Fe), amino acid and polyphenol content	Red-coloured, cloudy, sour, non-alcoholic	Black (purple) carrot ( <i>Daucus carota</i> ), turnip ( <i>Brassica</i> <i>rapa</i> )	Lactobacillus plantarum, L. br. delbrueckii, L. buchneri, Leucon cremoris, L. mesenteroides Pediococcus pentosaceus, Yeasts
Gilaburu juice	Antioxidants e.g. flavonoids, anthocyanins, ascorbic acid, and phenolic acids	Non-alcoholic and astringent without the addition of sugar	Cranberry bush ( <i>Viburnum opulus</i> L.)	Lactobacillus brevis, L. casei, L. p harbinensis, L. pantheris, L. pan pseudomesenteroides
Koumiss	Ash 0.4–0.5%, Ethanol 0.6–2.5%, CO2 0.5–0.9%, Lactose 3.5–4.3%, Lactic acid 0.7–1.8%, Protein 1.7– 1.9%, Lipid 0.6–1.3%, Total solid 10.6–11.3%, Ca, P, Mg	Milky gray colour, acidic, mildly alcoholic and sour, slightly carbonated fermented dairy beverage	Mare's milk or cow milk	Lactobacillus casei, L. bulgard fermentum, L. salivarius, L. bu plantarum, L. kefiranofacie Saccharomyces lactis, Streptococo

## Table 2 Summary of fermented traditional probiotic beverages of Turkish origin\*

\*Adopted and modified from [63

# Conclusion

Taking into account the numerous historical records describing the various health benefits that have been derived by man from the consumption of fermented foods, it goes without saying that probiotics have an ancient history, and particularly so with the Turkish and Turkic people. It can also be inferred that the long lifespan of people in prehistoric age is attributable partly or wholly to their regular consumption of health-promoting fermented probiotic products as encapsulated in this review. Furthermore, considering the many scientific researches that have been published worldwide over the years, there is sufficient data to show that probiotics are unarguably gaining more popularity than other functional foods. More importantly, it has been established in the current paper that Turkey is an origin country for several probiotic-containing functional beverages such as yogurt, kefir, boza, ayran, shalgam, hardaliye, gilaburu juice and koumiss, which have been concisely discussed in order to stir up deeper academic research interest. Owing to the fact that there is already more than enough data in the literature about yogurt globally, it has not been separately discussed in detail in the current review, rather the paper has focused on highlighting the other seven major beverages which seem not to have gained as much popularity as does yogurt. In conclusion, further research and publications especially aimed at demonstrating the medicinal health benefits of these indigenous Turkish beverages are recommended in order to promote more awareness about them and also encourage their habitual consumption by the populace.

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