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Physicochemical and Microbiological Properties of Kefir, Kefir Yogurt and Chickpea Yogurt

Nuray Güzeler, Elif Ari, Gözde Konuray, Çağla Özbek

***Abstract*—**The consumption of functional foods is very common. For this reason, many products which are probiotic, prebiotic, energy reduced and fat reduced are developed. In this research, physicochemical and microbiological properties of functional kefir, kefir yogurt and chickpea yogurt were examined. For this purpose, pH values, titration acidities, viscosity values, water holding capacities, serum separation values, acetaldehyde contents, tyrosine contents, the count of aerobic mesophilic bacteria, lactic acid bacteria count and mold-yeast counts were determined. As a result of performed analysis, the differences between titration acidities, serum separation values, water holding capacities, acetaldehyde and tyrosine contents of samples were statistically significant ($p < 0.05$). There were no significant differences on pH values, viscosities, and microbiological properties of samples ($p > 0.05$). Consequently industrial production of functional kefir yogurt and chickpea yogurt may be advised.

***Keywords*—**Chickpea yogurt, kefir, kefir yogurt, milk.

I. Introduction

MILK is a nutrient that has many ingredients for growing and developing of the organism and ensuring continuity of life [1]. The consumption of dairy products is very important because of the absence of alternative products [2]. Vitamin A, Vitamin D, calcium and phosphorus contents of milk are very high [3]. For this reason, milk and other dairy products are one of the most important foods that should be consumed. Drinking milk which is obtained by raw milk is at the first subgroup of dairy products. Yogurt and ayran are at the second subgroup and cheeses such as white cheese, Kasar cheese, Cokelek, Lor cheese (traditional Turkish cheeses) etc. are at the third subgroup. Butter is at the fourth subgroup, milk powder is at the fifth subgroup and lastly ice cream is at the sixth subgroup [4].

Yogurt is the most produced milk product subsequent to cheese [5] and the most demanded yogurt type is plain yogurt in Turkey. However, fruity and flavored yogurt types are generally preferred at western countries [6]. It is important to investigate dairy product varieties. These researches are generally about functional and natural products [7]-[12] Kefir yogurts and chickpea yogurts may be examples for these products. However further scientific researches about these are required.

According to Turkish Food Codex Fermented Milk Products Notification, kefir is a fermented dairy product and kefir culture ferments lactose by different strains of *Lactobacillus kefiri, Leuconostoc, Lactococcus* and *Acetobacter* [13]. The distinguishing property of kefir with other fermented dairy products is producing of it by kefir grains which are biologic and live organisms [14]. Kefir is a source of Vitamin B_1, Vitamin B_{12}, Vitamin K, calcium, amino acids, folic acid and biotin [15]. In the formation of kefir, microorganisms break down proteins in milk into peptone, peptide and aminoacids and lactose into lactic acid and alcohol. Final product can be easily digested [16]. The lactose content of kefir is lower than milk. For this reason, kefir may be a good solution for people who have lactose intolerance [17], [18]. In last years, kefir yogurt is produced by fermenting milk with kefir culture and incubating at 44 ℃. Kefir yogurt has taken place in the markets with name of Yofir which is produced in Güney Milk plant in the Mediterranean region of Turkey in 2016. The inoculum amount and incubation time are higher for this product.

Chickpea yogurt is another new functional and enriched milk product. For enrichment purpose, chickpea (*Cicer arietinum*), which is a legume of the family Fabaceae, subfamily Faboideae, is used [19]. The protein, mineral, vitamin and dietary fiber contents of chickpea are very important [20]. There is no scientific research about production of fermented milk product by directly using chickpea. However, there are some researches about fermented milk products that were produced by using chickpea extracts. In a research, chickpea which was kept in water at room temperature for one night is grounded. After this mixture was filtered, it was mixed with milk with 1:1 ratio and fermented with 6% of *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus.* Finally it was reported that using chickpea in yogurt production leads to obtain a product with high nutritional value [21]. In another research, enriched yogurt which has high nutritional value and biological quality was produced by adding chickpea extract to cow and camel milk. As a result of this research, it was reported that these yogurts may be evaluated as functional food in food industry [22]. The information about production of yogurt (fermented milk) by using chickpea was introduced at press and it became more common in last years. However there is no industrial production of this product [23]-[25].

In this research, kefir, kefir yogurt and chickpea yogurt were produced in three replications and their pH, titration acidities, viscosities, serum separation amounts, water holding capacities, acetaldehyde contents, tyrosine contents, aerobic mesophyll bacteria counts, lactic acid bacteria count, mold and

N. Güzeler is with Cukurova University Agricultural Faculty Food Engineering Department, Adana, Turkey (corresponding author, phone: +905325570179; e-mail: nsahan@cu.edu.tr).

E. Arı, G. Konuray, and Ç. Özbek are with Cukurova University Agricultural Faculty Food Engineering Department, Adana, Turkey (e-mail: ari.elif33@gmail.com, gkonuray@cu.edu.tr, cglaayldrm@gmail.com).

yeast count were analyzed.

II. Material and Method

A. Material

In this research, raw cow milk was supplied from Cukurova University Faculty of Agriculture, Research and Application Farm Animal Husbandry Branch in Turkey. Kefir grains, Kocbasi chickpea and milk powder were supplied from domestic markets. 200 ml of polypropylene yogurt bowls were used for packaging.

B. Method

For kefir production, milk was standardized and heated to 90 °C for 5 minutes. After it was cooled to 18-20 °C, 5% kefir culture was inoculated to milk. It was incubated until its pH value reached to 4.7. It was stored at 4 °C for 24 hours after incubation.

For kefir yogurt production, milk was standardized and 3% of milk powder was added to it. It was heated to 90 °C for 5 minutes and cooled to 40 °C. 5% of kefir culture was inoculated to milk. It was poured into the bowls and incubated until its pH value reached to 4.7. After incubation, it was stored at 4 °C for 24 hours.

For chickpea yogurt production, milk was standardized and 3% of milk powder was added to it. It was heated to 90 °C for 5 minutes and cooled to 44 °C. At the first step, 10 gram of chickpea was used for 200 ml of milk to obtain chickpea yeast. Then, the same procedure was replicated five times and a fermented milk product was produced. 5% of it was inoculated to milk. It was poured into the bowls and incubated until its pH value reached to 4.7. After incubation, it was stored at 4 °C for 24 hours. All productions were carried out in three replications and some physicochemical and microbiological properties of the samples were analyzed after 24 hours.

In this research, analysis of pH values [26], titration acidity values in terms of lactic acid (%) [27], protein contents [28], fat content according to Gerber method [27] were performed. The viscosity value measurements were performed by using Brookfield Digital Viscometer Model DVII+Pro [29]. The analysis of serum separation [30], [31], water holding capacity [32], acetaldehyde content according to iodimetric method [33], tyrosine content [34] was carried out. As microbiological analysis, the count of aerobic mesophyll bacteria [35], lactic acid bacteria count [36] and mold-yeast counts [37] were performed.

III. Results

The compositional values of raw milk that was used as raw material in the production are shown in Table I. As shown in Table I, the pH value of milk that was used in yogurt production was determined as 6.70, titration acidity in terms of lactic acid was 0.18%, protein content was 3.25% and fat content was 2.75%. According to Turkish Food Codex Raw Milk and Heat Treated Drinking Milk Notification; titration acidity of raw cow milk in terms of lactic acid should be between 0.135 and 0.2%, fat content should be at least 3.5% and protein content should be at least 2.8% [38]. Titration acidities and protein content of milk that was used in the production was appropriate for this scale. However, the fat content of it was lower. The reason of that may be seasonal differences, feeding style or age of the animal. Some physicochemical properties of kefir, kefir yogurt and chickpea yogurt are shown in Table II.

TABLE I
The Composition of Raw Milk

Properties	Raw milk
pH	6.70 ± 0.01
Titration acidity (L.a%)	0.18±0.02
Dry matter (%)	11.60±0.32
Protein (%)	3.25±0.05
Fat (%)	2.75±0.15

According to Turkish Food Codex Fermented Milks Notification; the titration acidities of kefir samples in terms of lactic acid should be at least 0.6%, titration acidities of yogurt should be between 0.6 and 1.5%. Titration acidities of kefir and kefir yogurt were found appropriate for the standard. The titration acidity of kefir yogurt was lower than kefir and chickpea yogurt. There were significant differences between titration acidities of kefir, kefir yogurt and chickpea yogurt samples ($p<0.05$).

Unlike titration acidity, pH values of kefir yogurt were higher than kefir and chickpea yogurt. The lowest pH value belonged to kefir samples. The differences between pH values of kefir, kefir yogurt and chickpea yogurt were found statistically significant ($p<0.05$).

The highest viscosity value which was measured at 15^{th} second was observed in chickpea yogurt. The viscosity of kefir yogurt was higher than kefir samples. At the 30^{th} second, similar results were observed. The highest viscosity value was shown in chickpea yogurt. Kefir yogurt and kefir followed it respectively. According to the statistical analysis, the differences between viscosity values of kefir, kefir yogurt and chickpea yogurt samples were not significant ($p>0.05$).

The highest serum separation amount was observed in kefir samples; kefir yogurt and chickpea yogurt samples followed it respectively. Serum separation is undesired situation for milk and milk products [39]. The differences between serum separation amount were found statistically significant on kefir, kefir yogurt and chickpea yogurt ($p<0.05$).

The highest water holding capacity was observed on chickpea yogurt. Kefir yogurt and kefir followed it respectively. There were statistically significant differences between water holding capacities of kefir, kefir yogurt and chickpea yogurt samples ($p<0.05$).

Acetaldehyde is the main and the most dominant flavor in yogurt [40]. The highest acetaldehyde content was observed on kefir, and the lowest acetaldehyde content was observed on chickpea yogurt. The reason of paucity of acetaldehyde content of chickpea yogurt may be caused by chickpea amount. These differences in the acetaldehyde contents of kefir, kefir yogurt and chickpea yogurt samples were

statistically significant ($p<0.05$).

TABLE II
SOME PHYSICOCHEMICAL PROPERTIES OF KEFIR, KEFIR YOGURT AND CHICKPEA YOGURT

Properties	Kefir	Kefir Yogurt	Chickpea Yogurt
pH	3.96±0.17[a]	4.27±0.27[a]	4.03±0.36[a]
Titration acidity (%L.a)	1.22±0.16[a]	0.99±0.26[b]	1.22±0.73[a]
Viscosity in 15 sec (cP)	1272.00±276.78[a]	1612.00±375.14[a]	1677.33±376.51[a]
Viscosity in 30 sec (cP)	1192.00±18.13[a]	1348.00±336.28[a]	1392.66±323.29[a]
Serum separation (%)	39.00±0.70[a]	37.99±1.68[ab]	36.11±0.70[b]
Water holding cap. (%)	36.81±0.60[b]	38.83±0.46[a]	39.50±0.50[a]
Acetaldehyde (ppm)	13.34±0.11[a]	8.85±0.08[b]	6.55±0.07[c]
Tyrosine (mg/g)	0.38±0.02[a]	0.30±0.02[b]	0.33±0.03[ab]

[a, b, c:] Values that are shown in the same line with different exponential letters are different in terms of $p<0.05$ level of significance.

As a result of proteolytic activities of yogurt cultures; peptides and amino acids revealed from proteins. Depending on amino acid level, taste disorders (rancidity) may occur in fermented milk products. Tyrosine content expresses total amino acid content as a result of proteolysis [41]. The highest tyrosine content was observed on kefir samples; chickpea yogurt and kefir yogurt followed it respectively. These differences were found statistically significant ($p<0.05$) and the reason of it may be fermentation time.

Microbiological properties of kefir, kefir yogurt and chickpea yogurt were shown in Table III.

TABLE III
MICROBIOLOGICAL PROPERTIES OF KEFIR, KEFIR YOGURT AND CHICKPEA YOGURT

Properties (log kob/ml)	Kefir	Kefir Yogurt	Chickpea Yogurt
The count of aerobic mesophill bacteria	7.45±2.33[a]	5.52±1.89[a]	4.91±2.88[a]
Lactic acid bacteria count	7.85±1.52[a]	6.35±0.74[a]	4.72±2.45[a]
Mold-yeast count	7.46±2.35[a]	5.52±1.83[a]	3.94±2.48[a]

[a, b, c:] Values that are shown in the same line with different exponential letters are different in terms of $p<0.05$ level of significance.

When the microbiological properties of samples were viewed, Kefir has the highest number of aerobic mesophill bacteria, lactic acid bacteria and mold-yeast. Kefir yogurt and chickpea yogurt samples followed it, respectively. The differences on microbiological properties of kefir, kefir yogurt and chickpea yogurt samples were not statistically significant ($p>0.05$).

IV. CONCLUSION

In this research; kefir, kefir yogurt and chickpea yogurt which are functional fermented dairy products were produced and their some physicochemical and microbiological properties were analyzed.

The pH values of obtained fermented products were lower and the titration acidities of them were higher than milk. The highest titration acidity value was observed on kefir and chickpea yogurt; the lowest one was observed on kefir yogurt. The highest viscosity value was observed on chickpea yogurt. The viscosity of kefir was lower than kefir yogurt. Serum separation amount of chickpea yogurt was found lowest and it was highest on kefir. The main flavor of yogurt, acetaldehyde content, was found highest in kefir and lowest in chickpea yogurt. The highest tyrosine content was found in kefir; chickpea yogurt and kefir yogurt followed it respectively. The highest bacteria counts were measured in kefir and lowest in chickpea yogurt.

According to the analysis results, there were statistically significant differences ($p<0.05$) in titration acidities, serum separation values, water holding capacities, acetaldehyde and tyrosine contents of samples. The differences of pH values, viscosity values and microbiologic properties were not found statistically significant ($p>0.05$).

Because of kefir yogurt and chickpea yogurt are new functional foods; there are not many scientific researches about these subjects. For this reason further researches are required.

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