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The Quality of Kefir with Honey and with Banana Enriched with Almond Milk

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ABSTRACT

Kefir is a fermented product obtained from goat, sheep and cow milk as a result of lactic acid and ethyl alcohol fermentation. In this study, it was aimed to investigate the possibility of producing an alternative functional kefir product from almond milk. It was enriched with banana and honey to improve its sensory properties. Samples were stored at +4°C for 14 days. The pH and dry matter of samples containing almond milk were lower than those containing cow's milk. Serum separation increased with the increase of almond milk ratio. The use of banana increased dry matter and viscosity, and decreased serum separation. pH, dry matter and viscosity decreased during storage. The highest L* value was observed in the control kefir produced from 100% cow's milk, and the lowest in the samples containing honey and banana on the 1st day of storage. It can be said that the addition of honey increases the b* value and the addition of banana decreases the b* value. As almond milk ratio and storage time increased, bacteria counts decreased. The lactococci counts of the samples with banana added (except for the control) were higher than the others. In the samples with banana and honey, a higher increase in yeast count was observed during storage compared to plain kefir (control sample). It can be said that the use of almond milk reduces the general acceptability of kefir. Almond milk can be successfully used in kefir production as a substitute for cow's milk, if sweeteners, fruit and thickeners are used.

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Introduction

Kefir is a valuable fermented milk product that is easily digested and obtained as a result of acid and alcohol fermentations (Karatepe and Yalçın, 2014). Kefir grains look like small cauliflower grains. Bacteria (lactobacilli, lactococci, *Leuconostoc* and *Acetobacter*) and yeasts in the structure of kefir grains give kefir a probiotic feature (Libudzisz and Piatkiewicz, 1990). Kefir has positive effects on the stomach, intestines and asthma. It is known to have many benefits such as reducing coronary heart diseases, strengthening the immune system, lowering cholesterol, regulating blood sugar, etc. In addition, it is antimicrobial, antitumor, anticarcinogenic and antiallergic (Karatepe and Yalçın, 2014). Industrial kefir production using kefir grain is quite difficult. The most suitable method in industrial kefir production is the use of starter culture, which will provide the desired properties instead of grain (Fontán et al., 2006). It has been stated that kefir produced by adding starter culture has a less acidic and creamy structure (Lopitz-Otsoa et al., 2006), more intense consistency and less yeasty flavor than the product produced with grain (Hafliger et al., 1991).

Unlike animal milks, plant milks contain significant amounts of phytochemicals (phenolic acids, flavonoids, stilbenes, lignans, hydrolyzable tannins, condensed tannins, proanthocyanidins, carotenoids, alkaloids, phytates, terpenes, phytoestrogens), dietary fiber, and have a low glycemic index (Chalupa-Krebzdak et al., 2018). In recent years, it is important both scientifically and commercially to obtain milk from some fruits that have proven many functional properties. These imitation milks can be produced by grinding and filtration of fruits after soaking, or by grinding raw-oiled or roasted fruits into flour without keeping them in water, and forming an emulsion by adding water (Bernat et al., 2014). Almond (*Prunus amygdalus*) is a tree species whose fruit can be eaten from the *Prunoideae* subfamily of the *Rosaceae* family. Almond is a fruit rich in protein, fat, carbohydrates and dietary fiber. It is useful for cardiovascular diseases due to the fatty acids and soluble fiber it contains, and it is good for anemia because it is rich in B group vitamins (B1, B2, B6). Because it is rich in calcium, it has a positive effect on bone and dental health. In addition, it is recognized as a protective functional fruit against hypertension due to its potassium-rich and low sodium content. When consumed regularly, it plays an important role in regulating blood sugar (Cassady et al., 2009). For vegetarian and vegan people who cannot consume milk and dairy products, products produced with probiotics added to plant-based milk will meet the needs of people who choose this diet (Erk et al., 2019).

For those who do not like to consume milk and its products, honey and various fruits can be added to milk and its products in order to improve their taste and smell. This can be true for plant milks as well. With the addition of honey and various fruits, the usefulness of the product is also increased. Studies have shown that sunflower honey (*Heianthus annuus*) is very rich in flavonoid content and has 42% of total phenolics (Amiot et al., 1989). Honey shows antioxidant and antimicrobial properties. Studies have also shown that the metabolites in the composition of honey have positive effects on the digestive system. It has been reported that the inhibitory effect of honey on cancer cells is caused by bioactive components such as phenolic acid and flavonoids in its structure, and these compounds prevent the formation of free radicals and oxidative stress that cause cancer (Mutlu et al., 2017). Since the sugars in honey can mix into the blood quickly, they provide energy and are easily digested. In addition, glucose, which is the energy source of the brain, increases the transport of tryptophan across the blood-brain barrier and is useful in the synthesis of serotonin, which has a function in brain work (Doğan, 2011).

Banana (*Musa sapientum, Musa paradisicia sapientum*) contains 70-75% water, 24-27% carbohydrates, 1% protein and 0.3% fat. In addition, 100 g of a banana contains about 900 IU/100 g of vitamin A, 0.4 mg of Vitamin B6, 0.6 mg of Vitamin B3, 0.3 mg of Vitamin B5, 9.8 mg of choline, 8.7 mg of vitamin C, 20 µg of folate, 27 mg of magnesium, 22 mg of phosphorus, 358 mg of potassium and 1 mg of sodium (Cemeroğlu, 1982; Ediboğlu Koç, 2019). The pH of banana is about 4.8; titratable acidity is 0.32 g/100 g (Hakim et al., 2012). Banana is loved by most people and preferred by athletes with the amount of potassium it contains. Also, it is a food that will relieve fatigue after training and fill the carbohydrate stores. Consumption of bananas before the training both ensures that the blood sugar remains in balance throughout the training and minimizes the mineral loss that will occur with sweating. In this way, it prevents cramps and muscle pain complaints after training. The fiber content of banana helps to keep the person full. Because of its fiber content, it regulates bowel movements. By increasing potassium and facilitating digestion in diseases with vomiting and diarrhea and electrolyte loss, it regulates the intestines, reduces the severity of symptoms and regulates blood pressure. It is very effective in protecting heart health by developing heart muscles. Potassium is also involved in the regulation of kidney activities. It prevents the formation of kidney stones. Therefore, banana is considered a kidney protective food. Thanks to the tryptophan amino acid it contains, bananas increase the hormones that give happiness and improve mood. This effect of banana definitely should be used in coping with depression and stress. The vitamins and minerals accelerate cell regeneration and make hair and skin tissues look lively and bright (Baykara, 2019). Banana also contains prebiotics such as inulin and oligofructose (Degeest et al., 2001; Flamm et al., 2001). 100 g of banana contains an average of 0.5 g of inulin and 0.5 g of oligofructose (Moshfegh et al., 1999).

The aim of this study is to produce kefir containing honey and banana from almond milk as an alternative functional product for vegetarians, those with lactose intolerance, the elderly, those with cardiovascular disease and diabetes, and to generate data for further studies in terms of its acceptability.

Materials and Methods

Materials

The liyophilized kefir starter culture (Doğadan Bizim Gıda ve Süt Ürünleri San. ve Tic. Ltd. Şti., Istanbul, Turkey) was used instead of kefir granules in this research. In the study, pasteurized cow's milk (Dost brand-daily milk, Bim United Mağazalar A. Ş., Turkey) and pasteurized almond milk (Kocamaar Tarım Ürünleri ve Tic. A. Ş., Mugla, Turkey) were used. The ripe banana used was obtained from the local market in Tekirdağ. As honey, filtered flower high plateau honey (Balparmak brand-Altıparmak Gıda San ve Tic A. Ş., Istanbul, Turkey) was used. 100 mL of almond milk used for making kefir contains 43 kcal of energy, 1.6 g of protein, 0.5 g of carbohydrates and 3.9 g of fat. 100 mL of pasteurized daily cow's milk contains 58.3 kcal of energy, 2.9 g of protein, 4.7 g of carbohydrates and 3.1 g of fat. The dry matter ratio of the honey is 84.55%, and the banana is 22.43%. The kefir culture includes *Lactobacillus kefir, Lactobacillus delbrueckii* ssp. *bulgaricus*, *Streptococcus thermophilus*, *Lactococcus lactis* ssp. *lactis*, *Lactococcus lactis* ssp. *cremoris*, *Lactococcus lactis* ssp. *lactis* biovar. *diacetylactis*, *Lactobacillus helveticus*, *Leuconostoc mesenteroides*, *Lactobacillus brevis*, *Lactobacillus casei*, *Lactobacillus plantarum*, *Kluyveromyces* sp. and *Saccharomyces* sp.

Methods

Production of kefir

In the production of kefir, cow's milk, almond milk and mixed in different proportions of cow's milk and almond milk are used. While some samples were containing honey, others contained both honey and banana: A: 100% pasteurized cow's milk [Control], B: 60% almond milk+ 25% pasteurized cow's milk + 5% honey+ 10% banana, C: 70% almond milk+ 25% pasteurized cow's milk + 5% honey, D: 85% almond milk + 5% honey + 10% banana, E: 95% almond milk + 5% honey, F: 100% almond milk. After inoculating 1% kefir into the milk brought to 25°C, fermentation continued until the pH decreased to 4.7 at 25°C (approximately 22 h). After completion of fermentation, kefir was cooled to 4°C and stored in glass bottles at 4°C for 14 days.

Physico-chemical analyses

pH measurements were made according to the electrometric method using a 300/310 branded WaterproofHand heldpH/mV/TemperatureMeter digital pH meter. The dry matter in kefir was calculated according to the gravimetric method. For serum separation analysis, 50 g kefir sample was weighed and kept at +4°C for 2 h, the amount of separated serum was determined in mL and the result was given as mL/50 g. Viscosity analysis was performed with the Sine-wave vibro Viscometer SV-10/SV-100 waveless vibrating fluidity meter. Konica-Minolta ChromaMeterCR-5 device was used for color analysis. L* (brightness), a* (+red, - green) and b* (+ yellow, - blue) values of kefir were examined.

Microbiological analyses

The counts of microorganisms were determined during storage at 4°C (1st, 7th and 14th days). One gram of sample was diluted with 9 mL of sterile 0.1% (w/v) peptone water (Oxoid, Basingstoke, UK), mixed with a vortex and subsequently serially diluted. The spread plate method was used to evalute of microbial counts. MRS agar (Oxoid CM 361, Thermo-Fisher Scientific Inc., USA) was acidified with HCl to reach 5.2 pH value. It was used to determine the count of *Lactobacillus* spp. and incubated anaerobically at 37°C for 72 h. M17 agar (Merck, Germany) was used for enumeration of *Lactococcus* spp. at 37°C for 48 h under aerobic conditions. Yeast counts were determined using Potato Dextrose Agar (Oxoid CM 139). Fermentation continued for 3-5 days at 25ºC. For total aerobic mesophilic bacteria count, incubation was carried out at 30°C for 48 h by inoculating with appropriate dilutions on Plate Count Agar (PCA) medium. After the incubation, plates were counted and results were expressed as log CFU/mL.

Sensory analyses

Kefir samples were evaluated in terms of color, appearance, structure, consistency, odor, taste, aroma, and general acceptability. For sensory evaluation, scores of 1, 2, 3, 4, and 5 were used for very bad, bad, average, good, and very good, respectively. Sensory evaluation in the study was carried out on day 1 of storage by 6 panelists.

Statistical analyses

In the statistical evaluation of the sensory analysis results, the difference between the samples was determined by applying one-way ANOVA analysis. In the statistical evaluation of physicochemical, color and microbiological analysis results, the difference between samples and days was determined according to the Random Plots Trial Plan. Analysis of variance was used to understand whether there was a difference between the results, and the "Duncan" multiple comparison test was used to determine the degree of difference. IBM SPSS Statistics21.0 (IBMCorp. USA) package program was used for statistical analysis.

Results

Physicochemical Properties

The pH values of kefir samples decreased during storage. The pH of samples D, E and F without cow's milk were lower than samples A, B and C with cow's milk. There was a decrease in the dry matter content of kefir samples during storage. The higher dry matter ratios of samples B and D compared to kefir produced from cow's milk can be associated with the 10% banana they contain. When the samples prepared with 100% cow's milk and 100% almond milk were compared, it was observed that kefir produced with cow's milk had more dry matter than those produced with almond milk. The highest (40%) serum separation was detected in the E sample containing 100% almond milk on the 1st day of storage. Serum separation increased with the increase of almond milk ratio. In the control sample, on the 7th and 14th days of storage, serum separation was not observed. While serum separation was not observed in the sample with high cow's milk and banana content, serum separation was considerably less in the sample with high almond milk and banana content compared to that produced from 100% almond milk. It was observed that the viscosity of kefir samples decreased during the storage period. The decrease in viscosities slowed after the first week of storage. The viscosity of samples D, E and F without cow's milk is lower than that of samples A, B and C containing cow's milk. The viscosity of the samples with the addition of banana is higher than that of the ones without the addition of banana. With the addition of banana, serum separation was also reduced due to the inulin in the structure of the banana (Table 1).

Color plays an important role in predicting the quality of the product before the consumer tastes it. On the 1st day of storage, the highest L* value was determined in control kefir, and the lowest L* value was determined in samples B and D containing honey and banana. During storage, the L* value decreased in the control kefir (from 14.59 to 4.68), while the others decreased until the 7th day and then increased. In general, a* values increased during storage. The highest a* values were detected in the C sample (70% almond milk + 25% cow milk +5% honey) on all storage days. The lowest a* values were also found in sample B (60% almond milk+ 25% cow milk+ 5%honey+ %10 banana). On the 1st day of storage, the highest b* value was determined in the control kefir, and the lowest b* value was determined in the B kefir sample. The b* value showed a continuous increase in sample D (D: 85% almond milk + 5% honey +10% banana) during storage. According to the results, it can be said that the addition of honey increases the b* value and the addition of banana decreases the b* value (Table 1). The time between peeling the banana and adding it to the milk, during which enzymatic browning can occur, can affect these values.

Microbiological Properties

On all days of storage, the control sample had the highest total mesophilic aerobic bacteria count, and F sample had the lowest total mesophilic aerobic bacteria count. As the ratio of almond milk increased, the total number of mesophyll bacteria decreased. It is thought that this result may be due to the chemical and microbiological differences between almond milk and cow's milk. Total mesophilic aerobic bacteria counts of kefir samples were generally lower on day 14 of storage than on day 1 of storage. Although the lactobacilli count of the control sample was higher on the 14th day of storage than on the 1st day of storage, lactobacilli counts decreased in all samples with almond milk added at the end of storage. At the end of storage, the highest lactobacilli count was detected in the sample produced from 100% cow's milk, while the lowest lactobacilli count was determined in the sample produced from 100% almond milk. It was determined that the highest number of lactococci at the beginning and end of the storage was in the A sample, the lowest in the E sample and generally decreased during the storage period. In general, lactococci counts in the banana added samples were higher than the lactococci counts in the other samples, except for the control sample. On the 1st day of storage, yeast counts of C and E samples (which do not contain bananas) were determined to be considerably higher than the other samples. While yeast counts decreased in these samples on the last day of storage compared to day 1, they increased in other samples (Table 2).

Table 1. Physicochemical properties of kefir samples during storage

Days	A	B	C	D	E	F
pH						
1	5.00±0.10 ^{E,c1}	4.73±0.03 ^{C,c}	4.84±0.03 ^{D,c}	4.72±0.02 ^{A,c}	4.70±0.04 ^{B,c}	4.65±0.05 ^{C,c}
7	4.76±0.01 ^{E,b}	4.55±0.05 ^{C,b}	4.72±0.01 ^{D,b}	4.35±0.05 ^{A,b}	4.45±0.03 ^{B,b}	4.55±0.05 ^{C,b}
14	4.72±0.02 ^{E,a}	4.50±0.05 ^{C,a}	4.62±0.02 ^{D,a}	4.33±0.03 ^{A,a}	4.39±0.04 ^{B,a}	4.50±0.04 ^{C,a}
Dry matter (%)						
1	10.65±0.05 ^{E,c}	11.35±0.01 ^{F,c}	10.30±0.01 ^{C,c}	10.79±0.09 ^{D,c}	8.30±0.10 ^{B,c}	6.21±0.06 ^{A,c}
7	10.40±0.10 ^{E,b}	10.98±0.03 ^{F,b}	9.54±0.02 ^{C,b}	10.16±0.01 ^{D,b}	8.00±0.10 ^{B,b}	4.15±0.01 ^{A,b}
14	10.27±0.01 ^{E,a}	10.30±0.01 ^{F,a}	9.61±0.01 ^{C,a}	9.98±0.02 ^{D,a}	7.14±0.03 ^{B,a}	3.76±0.01 ^{A,a}
Serum Seperation (%)						
1	8±0.10 ^{B,c}	0±0 ^{A,c}	0±0 ^{A,c}	6±0.00 ^{C,c}	40±0.02 ^{E,c}	34±0.00 ^{D,c}
7	0±0.00 ^{B,a}	0±0 ^{A,a}	0±0 ^{A,a}	4±0.10 ^{C,a}	20±0.50 ^{E,a}	16±0.20 ^{D,a}
14	0±0.00 ^{B,b}	0±0 ^{A,b}	0±0 ^{A,b}	6±0.05 ^{C,b}	36±0.01 ^{E,b}	30±0.02 ^{D,b}
Viscosity (mPa.S)						
1	7.60±0.01 ^{E,c}	11.50±0.05 ^{F,c}	6.56±0.02 ^{D,c}	5.88±0.04 ^{C,c}	3.35±0.05 ^{B,c}	2.95±0.05 ^{A,c}
7	5.84±0.04 ^{E,b}	6.00±0.10 ^{F,b}	4.69±0.04 ^{D,b}	3.56±0.10 ^{C,b}	2.23±0.03 ^{B,b}	2.06±0.06 ^{A,b}
14	5.18±0.08 ^{E,a}	5.27±0.02 ^{F,a}	3.94±0.05 ^{D,a}	3.44±0.04 ^{C,a}	2.21±0.01 ^{B,a}	2.04±0.04 ^{A,a}
Color						
L* value						
1	14.59±0.11 ^{D,c}	0.29±0.03 ^{A,c}	8.23±0.01 ^{C,c}	0.49±0.03 ^{B,c}	12.31±0.01 ^{E,c}	13.31±0.05 ^{F,c}
7	4.80±0.03 ^{D,a}	0.20±0.02 ^{A,a}	6.87±0.05 ^{C,a}	0.57±0.02 ^{B,a}	12.05±0.02 ^{E,a}	12.49±0.03 ^{F,a}
14	4.68±0.02 ^{D,b}	0.22±0.01 ^{A,b}	7.62±0.02 ^{C,b}	0.70±0.01 ^{B,b}	12.23±0.02 ^{E,b}	13.16±0.02 ^{F,b}
a* value						
1	0.87±0.05 ^{C,a}	1.06±0.05 ^{A,a}	6.41±0.04 ^{F,a}	1.54±0.07 ^{B,a}	4.89±0.03 ^{E,a}	3.41±0.04 ^{D,a}
7	3.92±0.07 ^{C,b}	0.86±0.02 ^{A,b}	6.42±0.06 ^{F,b}	1.74±0.04 ^{B,b}	4.90±0.02 ^{E,b}	3.79±0.03 ^{D,b}
14	4.45±0.04 ^{C,c}	1.04±0.05 ^{A,c}	6.70±0.02 ^{F,c}	2.10±0.06 ^{B,c}	4.91±0.02 ^{E,c}	3.97±0.02 ^{D,c}
b* value						
1	23.03±0.13 ^{D,c}	0.42±0.04 ^{A,c}	13.35±0.02 ^{C,c}	0.74±0.03 ^{B,c}	17.71±0.02 ^{F,c}	16.02±0.03 ^{E,c}
7	7.94±0.03 ^{D,a}	0.27±0.04 ^{A,a}	11.24±0.06 ^{C,a}	0.88±0.05 ^{B,a}	17.31±0.06 ^{F,a}	15.28±0.04 ^{E,a}
14	7.85±0.02 ^{D,b}	0.27±0.01 ^{A,b}	12.33±0.04 ^{C,b}	1.14±0.06 ^{B,b}	17.18±0.04 ^{F,b}	15.38±0.05 ^{E,b}

A: 100% cow milk (Control); B: 60% almond milk+ 25% cow milk+ 5%honey+ %10 banana; C: 70% almond milk + 25% cow milk +5% hone y; D: 85% almond milk + 5% honey +10% banana; E: 95% almond milk + 5% honey; F: 100% almond milk; [1]Upper case shows difference between samples, lower case shows difference between storage days ($P<0.01$)

Table 2. Microbiological properties of kefir samples during storage

Days	A	B	C	D	E	F
Total mesophilic aerobic bacteria number (log cfu ml^{-1})						
1	8.85±8.00 ^{D,c1}	8.08±7.30 ^{C,c}	8.11±7.78 ^{B,c}	8.04±7.00 ^{C,c}	7.66±6.30 ^{AB,c}	7.58±6.00 ^{A,c}
7	8.70±7.70 ^{D,b}	7.57±6.30 ^{C,b}	7.53±6.00 ^{B,b}	8.17±7.67 ^{C,b}	7.32±6.58 ^{AB,b}	7.38±7.18 ^{A,b}
14	8.48±7.30 ^{D,a}	8.11±7.30 ^{C,a}	7.31±7.00 ^{B,a}	7.90±6.00 ^{C,a}	7.40±6.70 ^{AB,a}	7.10±6.66 ^{A,a}
Lactobacilli number (log cfu ml^{-1})						
1	8.25±7.40 ^{C,c}	8.59±8.31 ^{B,c}	9.96±7.02 ^{A,c}	8.34±7.90 ^{B,c}	7.81±7.52 ^{A,c}	8.02±6.30 ^{A,c}
7	8.70±7.90 ^{C,b}	7.89±7.27 ^{B,b}	7.54±6.70 ^{A,b}	8.11±7.42 ^{B,b}	7.50±6.88 ^{A,b}	7.50±6.79 ^{A,b}
14	8.38±7.30 ^{C,a}	8.11±7.30 ^{B,a}	7.47±6.96 ^{A,a}	7.95±6.30 ^{B,a}	7.41±6.76 ^{A,a}	7.25±6.78 ^{A,a}
Lactococci number (log cfu ml^{-1})						
1	9.70±9.00 ^{C,c}	9.25±8.30 ^{B,c}	8.49±7.42 ^{A,c}	7.62±6.30 ^{A,c}	7.70±1.00 ^{A,c}	7.40±0.00 ^{A,c}
7	8.60±7.48 ^{C,b}	7.79±7.53 ^{B,b}	7.48±6.70 ^{A,b}	8.12±7.46 ^{A,b}	7.45±7.03 ^{A,b}	7.44±6.69 ^{A,b}
14	8.12±7.18 ^{C,a}	8.08±7.30 ^{B,a}	7.16±6.58 ^{A,a}	7.70±7.00 ^{A,a}	6.28±6.98 ^{A,a}	7.10±6.81 ^{A,a}
Yeast number (log cfu ml^{-1})						
1	2.04±1.00 ^{A,b}	2.03±0.76 ^{A,b}	4.60±3.18 ^{A,b}	2.00±0.00 ^{A,b}	6.30±5.30 ^{B,b}	2.00±0.00 ^{A,b}
7	2.70±1.00 ^{A,a}	3.04±2.00 ^{A,a}	4.78±4.00 ^{A,a}	3.97±2.30 ^{A,a}	3.40±2.00 ^{B,a}	2.70±1.00 ^{A,a}
14	3.01±1.76 ^{A,a}	4.19±3.61 ^{A,a}	4.48±0.00 ^{A,a}	4.07±3.86 ^{A,a}	5.18±4.00 ^{B,a}	3.70±3.00 ^{A,a}

A: 100% cow milk (Control); B: 60% almond milk+ 25% cow milk+ 5%honey+ %10 banana; C: 70% almond milk + 25% cow milk +5% hone y; D: 85% almond milk + 5% honey +10% banana; E: 95% almond milk + 5% honey; F: 100% almond milk; [1]Upper case shows difference between samples, lower case shows difference between storage days ($P<0.01$)

Sensory Properties

Control kefir had the highest score (4.8) in terms of color and appearance, and sample B had the lowest score (3.7). The structure and consistency scores of kefir samples vary between 3.17 and 4.7. The sample A had the highest score in terms of structure and consistency, and sample E had the lowest score. The sample B had the highest score (4.83) in terms of smell whereas the sample E had the lowest score (3.3). Taste and aroma scores range from 2.7 to 4.17. Samples A and B received the highest scores in terms of taste and aroma, while sample E received the lowest score. The sample A had the highest score (4.7) in terms of general acceptability, followed by samples B, C, D, E and F, respectively. It can be said that the use of almond milk reduces the general acceptability of kefir. As the ratio of almond milk in kefir increased, taste and aroma scores decreased (except for the sample F). This result is thought to be related to the unique aroma of almond. In kefir samples prepared with almond milk, a more intense aroma was felt compared to kefir prepared with daily milk. The general acceptability of kefir decreased with increasing almond milk content. The results show parallelism with the results of sensory evaluation. According to the results of the statistical analysis, the effect of the difference between the taste and aroma values and the general acceptability values of the samples was found to be significant at the $P<0.05$ level. The effect of the difference between samples on other evaluation criteria was found to be statistically insignificant ($P>0.05$). According to Duncan test results, A and B samples were similar in terms of taste and aroma. E sample is different from these. Samples C, D and F showed statistically similar characteristics with samples A, B and E. According to Duncan test results, C, D, E and F samples were found to be statistically similar in terms of general acceptability. Sample A is different from these. Sample B is statistically similar to all other samples (Figure 1). It is thought that the lower scoring of the B sample in terms of color and appearance compared to the A sample may be due to the color darkening due to the enzymatic browning reaction in the banana. It can be said that as the ratio of almond milk in kefir increases, the structure and consistency scores decrease. This result can be attributed to the content of almond milk. The viscosity values we obtained are in parallel with the sensory evaluation results.

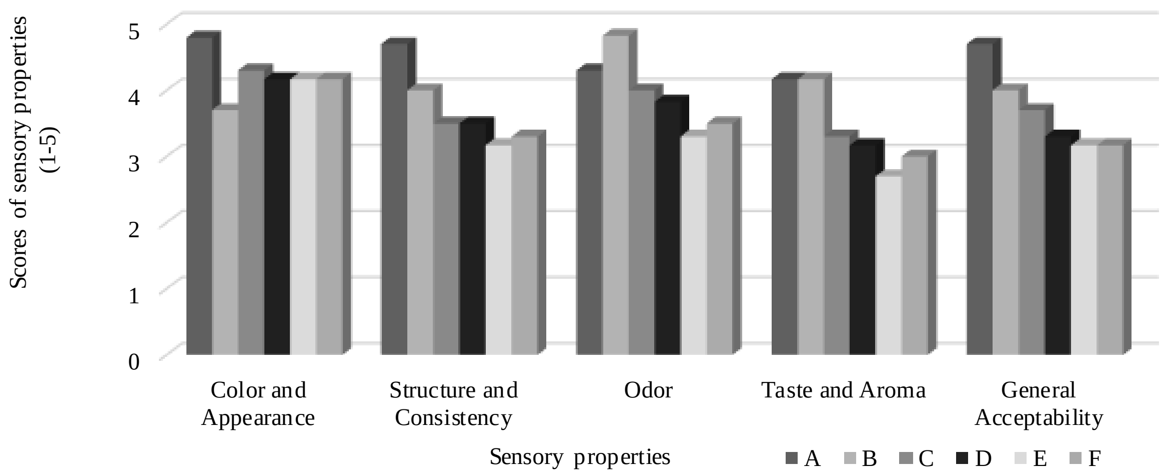

Figure 1. Sensory properties of kefir samples
A: 100% cow milk (Control); B: 60% almond milk + 25% cow milk + 5%honey + %10 banana; C: 70% almond milk + 25% cow milk +5% honey; D: 85% almond milk + 5% honey +10% banana; E: 95% almond milk + 5% honey; F: 100% almond milk

Discussion

Physicochemical Properties

The pH of fermented almond milk produced by Bernat et al. (2015) using *L. reuteri* ATCC 55730 and *S. thermophilus* CECT 986 is 4.67 on the 1st day of storage. It did not show any significant change until the 14th day. This value is similar to day 1 values in our study. In our study, the diversity of microorganisms is high due to the fact that the product is kefir. Therefore, the pH drop during storage was greater than theirs. In a study, the pH of kefir containing 2% inulin produced from cow's milk was lower than the pH of control kefir during storage. Since the banana used in this study also contains inulin, microorganism growth may have been positively affected and the pH value may have been lower than the samples that did not contain bananas (Tratnik et al., 2006). In Doğan (2011)'s study, the pH values of 10%, 20% and 30% flower honey kefir samples were lower than the pH of the control kefir sample. As the honey content increased, the pH decreased. In this study, the pH values of honey kefir samples were lower than the pH of the control sample. In the study of Topçuoğlu (2019), the pH of yogurt produced with cow's milk during storage decreased more than the pH of yogurt produced with almond milk. Similar results were obtained in this study as well. Koca (2016) produced kefir yogurt with banana after incubation at 39 ^{o}C using cow's milk, 5% sugar and 7% banana and stored the yogurts for 14 days. The pH values of the banana samples were lower than the plain ones. It was determined that the pH values decreased during 14 days. The results are similar to those in this study. In the study of Harmankaya et al. (2019) kefir milk was fermented after adding 2% kefir grains and 20% banana, and then stored for 14 days. As in the present study, in their study, the pH of banana kefir was lower than that of plain kefir during storage.. In addition, in the present study, the use of honey and almond milk in banana kefirs caused a further decrease in pH value.

The dry matter of the yogurt sample prepared by Bakirci and Kavaz (2008) with the addition of banana and sugar increased with the addition of banana. The amount of dry matter decreased during the storage period. The decrease in dry matter during storage may be due to the breakdown of proteins.

In one study, serum separation was found between 3.45– 4.95 mL/25 g in banana probiotic yogurts and between 5.27 and 5.88 mL/25 g in plain probiotic yogurts (Kavaz, 2006). This may be due to the serum retention of the mesh structure of the banana added to the yogurt (Çakmakçı et al., 1997). In this study, the serum separation of the samples with the addition of banana was low. Serum separation of fermented almond milk produced by Bernat et al. (2015) on the 1st day of storage is 43%. It decreased to 39% on the 14th day of storage. As the almond ratio increased, serum separation increased, and the ratio decreased with storage. In this study, no serum separation was observed in kefirs containing cow's milk. It was observed that as the ratio of almond milk increased, the rate of serum separation increased. In the study conducted by Yılmaz Ersan and Topçuoğlu (2019), the rate of serum separation increased as the almond milk ratio increased in yoghurts produced from cow's milk and almond milk. Due to the water-holding properties of milk proteins and milk fat globules, serum separation in the control sample was lower during storage than in other samples. In previous studies with plant milks, stability problems were observed due to the low content of proteins that act as emulsifiers in water-oil emulsions (Walstra, 1983). This problem is usually solved by adding hydrocolloids such as xanthan gum, changing the solvent properties of the aqueous phase depending on a modification of pH and forming a gel by increasing hydrogen bonds (Bernat et al., 2014; Yılmaz Ersan and Topçuoğlu, 2019]. Machado et al. (2017) stated that casein micelles in yogurt absorb water and reduce the release of water to the environment. They reported that although honey is a high-viscosity fluid, when the product is kept cold, it behaves like a pseudo-plastic fluid and provides better resistance to yogurt. In this study, while serum separation was not observed in the samples containing cow's milk and honey, the serum separation of the sample containing almond milk and honey was higher than the serum separation of the sample prepared with almond milk alone. This may be due to the fact that honey did not show the expected effect in the sample produced from casein-free almond milk.

In the study of Tratnik et al. (2006), the viscosity of kefir produced with cow's milk was found to be 101.1, 89.4 and 75.1 MPa s on the 1st, 5th and 10th days of storage, respectively. The viscosity of kefir with 2% inulin added was found to be 121.6, 91.4 and 78.8 MPa s on the 1st, 5th and 10th days of storage, respectively. Yaygın (1999) reported that pectin in the structure of the fruit swells, causing an increase in consistency, that is, increasing the viscosity. It is seen that this result is similar in samples B and D. In the study of Doğan (2011), the viscosity of kefir samples containing honey was lower than the viscosity of the control kefir sample. As the honey ratio increased, the viscosity decreased. In this study, the viscosity of the kefir sample (C) containing honey is lower than the viscosity of the control sample (A) (Table 1).

Karaca et al. (2013) reported that as the fruit fiber content increased, the L* value decreased and the highest L* value was seen in control yogurts without added fruit fiber. In the study of Doğan (2011), kefir with flower honey was produced using cow's milk, in the control sample the L*, a* and b* values were 73.18, -1.35 and 3.77, respectively; while the L*, a and b values of kefir with 10% honey added were 71.18, -1.50 and 5.22 respectively. It has been reported that there is a very close relationship between the phenolic components of honey and the color of honey, and the color darkens with the increase of phenolic components (Can, 2014). Since a high L value indicates a light color and a low L* value indicates a dark color, it is expected that the L* value will be low in honey added samples. Machado et al. (2017), a * and b * values increased with the increase in the amount of honey added in 7-day storage, as in this study. In the study of Koca (2016), in which she investigated kefir yogurts with the addition of banana, L*, a* and b* values decreased towards the 7th day of storage and increased towards the 14th day of storage, as in this study.

Microbiological Properties

Bernat et al. (2015) determined that there was a decrease in the number of these bacteria during storage in the fermented milk product they prepared using almond milk and *L. reuteri* and *S. thermophilus*. In a study conducted by Erdoğan et al. (2019), it was reported that the *Lactobacillus* spp. number of kefirs produced from cow's milk using kefir grains was 10.54 log CFU mL^{-1}, and the *Lactobacillus* spp. number of kefirs produced using kefir starter culture was 8.40 log CFU mL^{-1}. Starter culture was also used in this study and their results are similar to the control sample in this study. In a study by Kök-Taş et al. (2013), the *Lactobacillus* spp. count of kefir produced from cow's milk using starter culture was 9.27 log CFU mL^{-1}, 9.26 log CFU mL^{-1}, 9.06 log CFU mL^{-1} and 8.89 log CFU mL^{-1} on the 1st, 7th, 14th and 21st days of storage, respectively. It has been reported that the number of *Lactobacillus* spp. decreased during storage. Topçuoğlu (2019) found *L. bulgaricus* numbers varied between 7.90 and 9.48 CFU g^{-1}, and the number of lactobacilli decreased as the almond milk ratio increased. Their result is in agreement with that in this study.

In the study of Harmankaya et al. (2019), the number of lactobacilli on the 1st day of storage was determined as 7.81 log CFU mL^{-1} in plain kefir and 6.98 log CFU/mL^{-1} in banana kefir. These numbers increased to 8.00 and 8.04 on the 14th day of storage for plain and banana kefir. In the present study, the lactobacillus count of plain kefir made from cow's milk increased during storage. However, the number of lactobacilli decreased during storage in kefir supplemented with almond milk. The lactobacilli count of kefirs with banana added with high almond milk ratio was slightly higher than the others.

In the study of Harmankaya et al. (2019), the lactococcal count on the 1st day of storage was determined as 9.08 log CFU mL^{-1} in plain kefir and 8.48 log CFU mL^{-1} in banana kefir. These numbers decreased to 6.68 and 7.30 log CFU mL^{-1} in plain and banana kefir on day 14 of storage. Yılmaz Ersan and Topçuoğlu (2019) produced yogurt from a mixture of cow's milk and almond milk. They determined that the number of *S. thermophilus* in yoghurts with high cow's milk content was higher than yoghurts with high almond milk content, and its number decreased with storage. Similar results were found in this study as well.

It was observed that the yeast counts in kefir produced from cow's milk and almond milk were close to each other. In the samples of kefir with banana and honey, there was a higher increase in the number of yeasts during storage compared to plain kefir. The addition of carbohydrates along with the addition of bananas may have caused an increase in the number of yeast. In the study of

Harmankaya et al. (2019), while the yeast count of plain kefir produced from cow's milk decreased by 1.247 log CFU mL^{-1} during 14 storage days, the yeast count in kefir with banana added decreased by 0.46 log CFU mL^{-1} during this period. Yeast counts were higher in C and E samples, which did not contain bananas but contained honey, compared to the other samples. This is thought to be due to yeast fermenting sugar vigorously.

Tamime et al. (2005) stated that for probiotic foods to have a positive effect on health, the product should contain at least 10^6 CFU g^{-1} of live microorganisms during storage, and the daily amount to be taken in the product should be 10^8 -10^9 CFU g^{-1} for the expected therapeutic effect to be seen. In the "List of Health Claims Except for Statements Regarding Reducing the Risk of Disease, Development and Health of Children" in the Turkish Food Codex Regulation on Nutrition and Health Claims, it is stated that in order for food to be considered probiotic, it must contain at least 1.0×10^6 CFU g^{-1} live probiotic microorganisms (TGK, 2017). The number of microorganisms determined in this study is well above the specified values.

Sensory Properties

Sensory properties of milk and dairy products provide important information about their quality (Ünal and Besler, 2008). In a study, Uslu (2010) examined a total of 33 samples consisting of plain, fruity and diet kefir produced from cow's milk in 5 different companies in Ankara. As a result of sensory analysis, fruit-mixed and banana kefirs were more appreciated than plain ones. In the study of Harmankaya et al. (2019), it was determined that banana kefir had the best appearance, consistency and smell, while plain kefir had the best flavor. In the overall evaluation, the panelists liked the banana kefir the most, followed by plain, strawberry and apricot kefir. In our study, kefir with the highest pH value (plain cow's milk and banana) got the highest score in terms of viscosity and was generally appreciated more by the panelists. Özer and Atamer (1994) attributed this situation to the decrease in environmental acidity as the pH level increased and the decrease in the denaturation rate to affect the viscosity.

Conclusion

Kefir can be produced by using almond milk as an alternative functional product for vegetarians, lactose intolerant people, people with cardiovascular disease and diabetes. Increasing consumption of almond will provide advantage in terms of health. With this study, it was determined that some studies should be done to improve the sensory and structural properties of kefir to be produced with almond milk. The use of sweetener and fruit will be positive for improving the taste and smell of kefir, which will be produced from almond milk. Honey or an herbal sweetener may be suitable as a sweetener. Herbal thickeners can be used to increase viscosity and reduce serum separation. Probiotic almond drinks could create potential for this market. The results of the analysis showed that almond milk can be successfully used as a substitute for cow's milk in kefir production.

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