Physicochemical and Rheological Properties of Commercial Kefir Drinks

Oguz Gursoy1, Kübra Kocatürk2, Hande Özge Güler Dalı1, Hasne Nazlı Yakalı2, Yusuf Yilmaz1

1Department of Food Engineering, Faculty of Engineering and Architecture, Burdur Mehmet Akif Ersoy University, İstiklal Campus, TR-15030, Burdur, Turkey
2Division of Food Engineering, Graduate School of Natural and Applied Sciences, Burdur Mehmet Akif Ersoy University, İstiklal Campus, TR-15030, Burdur, Turkey

ABSTRACT

Kefir is one of the most preferred fermented beverages, and its production constitutes a significant portion of the dairy industry. In this study, the physicochemical and rheological properties of commercial plain kefir drinks were compared. Kefir drinks of dominant brands in the Turkish kefir market (n=6) were obtained from national markets, and samples with different production dates for each brand were used as replications. The ranges for the dry matter, protein and fat contents (%) of kefir drinks were 9.49-11.97, 2.30-3.44 and 2.50-3.00%, respectively. Moreover, the pH values of samples were in a range between 3.86 and 4.06 while their total titratable acidity (% lactic acid) values ranged from 0.71 to 0.93%. Significant differences were found among the physicochemical properties of commercial kefir drinks (p<0.05). Rheological analyses were conducted at three different temperatures (4.0, 10.0 and 25.0±0.3°C), and the power law model was used to interpret the rheological characteristics of kefir drinks, which resulted in high coefficient of determination (R²) values. Besides, the apparent viscosity values of samples (at a shear rate of 111.6 s⁻¹) were inversely related to temperature and varied between 0.48 and 1.76 mPas (p<0.05). Activation energy values of samples were between 21.51 and 36.18 kJ/mol. Significant differences in physicochemical and rheological properties were found among the commercial kefir samples of different brands, indicating the compositional variations of raw materials and kefir cultures used in their production.

Keywords: Kefir, Fermented milk, Consistency, Rheological behavior, Viscosity

ÖZ

Kefir, fermentli içecekler içerisinde en çok tercih edilenlerden biri olup üretim süs endüstrisinde önemli bir yer tutmaktadır. Bu çalışmada, ticari sade kefirlerin fizikokimyasal ve reolojik özellikleri karşılaştırılmıştır. Türkiye ticari kefir pazarında etkili olan markalara ait kefirler (n=6) Türkiye'deki ulusal marketlerden temin edilmiş ve her bir marka için farklı son kullanma tarihlerine sahip örnekler tekrarır olarak kullanılmıştır. Ticari kefir örneklerinin kuru madde, protein ve yağ içeriklerine ait araçıklar, sırasıyla 9.49-11.97, 2.30-3.44 ve 2.50-3.00 olarak bulunmuştur. Örneklerin pH değerleri 3.86-4.06 aralığında, toplam titre edilebilir asitlik (% , laktik asit) değerleri %0.71-0.93 arasında saptanmıştır. Farklı markalara ait ticari kefir örneklerinin fizikokimyasal özelliklerinde önemli değişiklikler saptanmıştır (p<0.05). Farklı sıcaklıklarında (4.0, 10.0 ve 25.0±0.3°C) yapılan reolojik analizlerde, kefir özelliklerinin reolojik özelliklerinin yorumlanmasında yüksek R² değerlerinin elde edildiği Üssel Kural (Power Law) modeli kullanılmıştır. Örneklerin (111.6 s⁻¹) kayma oranındaki) görünür viskozite değerlerinin sıçaklıkla ters orantılı olduğu ve 0.48 ile 1.76 mPas arasında değiştiği saptanmıştır (p<0.05). Örneklerin aktivasyon enerjisi değerleri 21.51 ve 36.18 kJ/mol arasında değişmiştir. Farklı markalara ait ticari kefir örneklerinin fizikokimyasal ve reolojik özellikleri arasında önemli
INTRODUCTION

Commercial production of probiotic foods has greatly increased over the last decades in parallel to an increase in consumer consciousness towards healthy food choices. Fermented milk beverages are particularly effective in preserving the diversity of microorganisms and balance in the intestinal flora by their probiotic content. Kefir is a traditional fermented milk drink with an increasing popularity, and it can regulate the intestinal microflora by supplementing a number of probiotic bacteria that are useful for maintaining the body health [1, 2].

Traditionally produced kefir is highly preferred by consumers, and commercial producers of kefir drinks have a target to obtain taste and aroma similar to those of traditional kefir drinks. In kefir production, thermophilic, mesophilic, probiotic bacteria and yeasts are mostly used together or separately as a kefir culture, which gives unique flavor to the product by their symbiotic metabolic activities. The type and ratio of starter used in kefir production are effective on the sensory and chemical qualities of the final product [3-5]. In commercial production, kefir is produced by microorganisms in the form of kefir grains that resemble cauliflower florets or by isolated microorganisms as a starter culture. Kefir has an important functional property with its characteristic microbial flora, flavor and slightly acidic sour taste, in addition to its appetizing and refreshing characteristics. While ethyl alcohol, acetaldehyde, diacetyl and amino acids, which contribute to the sensory properties of kefir, are formed during fermentation, the primary products of this process are ethanol, lactic acid and carbon dioxide that characterize sour, acidity and low alcoholic taste, also viscous and foamy texture of kefir drinks [1, 6]. The health benefits of kefir intake may include an improvement in lactose tolerance and immune system, an increase in anticarcinogenic and antimutagenic properties, sensorial and nutritional attributes of milk beverage [4, 7, 8].

Rheological characteristics of kefir influence its consumer acceptability and choice directly. Various factors have been reported to be effective on the formation of the main characteristics and composition of kefir depending on the raw material used such as type of milk and diversity of microorganisms, also process and final product variations as fermentation time, temperature, and storage time or flavor addition [9-11].

To the best of our knowledge, studies examining the rheological properties of kefir samples at different serving temperatures are quite limited. In one of these studies, the viscosity values of commercial kefir samples (n=6) were reported at 50 and 100 rpm without any temperature information [12]. In another study, the rheological properties of a commercial kefir sample were determined at 10, 20 and 30°C, which included temperatures beyond the regular consumption temperatures of kefir drinks [13]. In this present study, the rheological properties of six different commercial kefir samples were determined at 4, 10 and 25°C and the effects of different serving temperatures on the rheological behavior of commercial kefir samples were evaluated.

MATERIALS and METHODS

Materials

Commercial kefir drinks of the dominant brands in Turkey were purchased from various markets, and samples with different production dates were used as a replicate. Then, they were coded as A, B, C, D, E and F and kept at 4°C prior to analyses.

Physicochemical Analyses

The dry matter contents of kefir samples were determined by a rapid moisture analyzer (Kern DBS 60-3, Balingen, Germany) at 110°C, and pH values were determined by a pH meter (Jenco 6173, Jenco, San Diego, CA, USA). Color parameters (L*, a* and b* values) were measured in the CIELAB system by a colorimeter (CR-400 Chroma Meter, Konica Minolta, Japan). During measurements, illuminant D65 and a pulsed xenon lamp as a source of light were used in the device. Before the analyses, the device was calibrated with its white calibration plate then CIE L*, a*, and b* color parameters were obtained. The titratable acidity values of kefir samples were determined according to the method of Oysun [14]. Total nitrogen analyses were conducted by the Dumas method with the Dumas analyzer (Gerhardt GmbH & Co. KG, Königswinter, Germany), and the protein content of kefir samples was determined by using the conversion factor of 6.38. The fat content of kefir samples was determined by the Gerber method [14].

Rheological Analyses

The rheological properties of kefir samples were determined by a Brookfield viscometer (Model DV-II Pro, Brookfield Engineering Laboratories, Middleboro, MA, USA) with a spindle of RV-2. For this purpose, a portion of the sample (~500 mL) was placed into a 600 mL beaker, which was carefully isolated by a Styrofoam to minimize heat loss during measurements, and measurements were carried out at a constant temperature. Each measurement was repeated 5 times at the shear rates of 40-120 rpm (37.2-111.6 s⁻¹) for each temperature (4, 10 and 25°C). Shear rate (s⁻¹), shear stress (dyn/cm²), torque (%) and viscosity (cp) values were measured at intervals of 10 s. The apparent
viscosity values of commercial kefir samples were determined at 120 rpm (111.6 s'). Apparent viscosity and torque values were recorded for each shear rate, and the flow behavior index values (n) and consistency coefficients (K, Pa.s^n) of kefir samples were determined according to the power law model (Equation 1), where δ and γ demonstrate the shear stress (Pa) and the shear rate (s^-1), respectively [15]. The Arrhenius equation (Equation 2) was used to determine the temperature dependency of apparent viscosity and to calculate the activation energy values according to the method of Marcotte et al. [16]. In Equation 2, η is apparent viscosity (Pa.s), A is the frequency factor, E_a, R, T are activation energy, gas constant and temperature (K), respectively.

\[ \delta = K(\gamma)^n \]
\[ \eta = A e^{(-E_a/(RT))} \]

Statistical Analyses

Statistical analyses were conducted using the variance analysis (ANOVA) of Statistical Analysis Software (SAS® Package Version v9.3 (SAS® Institute, 2010). Mean values of the analyses were compared for significant differences by the Duncan’s multiple-range test with the significance level of 0.05. All analyses were conducted in duplicates, and results were expressed as a mean ± standard deviation. PROC CORR procedure was used to obtain the Pearson’s correlation of physicochemical and rheological properties of samples in 95% confidence interval.

RESULTS and DISCUSSION

Physicochemical Properties

The chemical analysis results of commercial kefir drinks are presented in Table 1.

| Sample code | Dry matter (%) | Protein (%) | Fat content (%) on dry matter basis | Fat (%) | pH | Total titratable acidity (% Lactic Acid) |
|-------------|----------------|-------------|------------------------------------|---------|----|----------------------------------------|
| A           | 11.71±0.22^a   | 2.63±0.09^c | 26.04±0.06^BA                      | 3.00±0.00^A | 4.06±0.04^A | 0.81±0.02^B |
| B           | 9.96±0.19^e    | 2.61±0.09^c | 28.49±1.39^A                       | 2.85±0.07^BAC | 3.90±0.05^C | 0.80±0.02^B |
| C           | 9.49±0.10^f    | 2.30±0.16^c | 26.13±0.04^A                       | 2.50±0.00^C  | 3.88±0.03^C | 0.91±0.02^A |
| D           | 10.51±0.07^d   | 3.06±0.29^b | 27.49±2.68^A                       | 2.90±0.28^BA | 3.88±0.04^C | 0.93±0.02^A |
| E           | 11.01±0.07^c   | 3.44±0.11^a | 22.76±0.22^BC                      | 2.50±0.00^C  | 3.97±0.02^B | 0.71±0.04^C |
| F           | 11.97±0.07^a   | 3.24±0.37^BA| 21.34±1.99^C                       | 2.55±0.21^BC | 3.86±0.02^C | 0.89±0.01^A |

*Values with different letters in a column indicate statistical significances among means (p<0.05)

The dry matter, protein and fat contents (%) of commercial kefir samples were in the intervals of 9.49±0.10-11.97±0.07%, 2.30±0.16-3.44±0.11%, and 2.50±0.00-3.00±0.00%, respectively. Moreover, pH and total titratable acidity (%, lactic acid) values of the samples were determined in the intervals of 3.86-4.06 and 0.71-0.93%, respectively. The pH values of the brands A and E were higher than the pH values of other brands (p<0.05), and insignificant differences were determined in pH values of other four brands. Variations in other physicochemical properties were obtained among different sample groups (p<0.05). For the protein contents of kefir drinks, brands A, B and C were similar (p>0.05) while the protein content of brand E was significantly higher compared to other brands with an exception of brand F (p<0.05). Also, significant differences determined in dry matter values of all six brands that the dry matter content of brand F was significantly higher than others (p<0.05), while the lowest dry matter content value was obtained for brand C (p<0.05). Moreover, an insignificant difference in fat contents (%) was found among A, B and D brands (p>0.05) and also among C, E and F brands (p>0.05). Similar results were reported by Barukčić et al. [17] who compared chemical, sensory and rheological properties of kefir produced by different combination of kefir starters and grains. In their study, pH and titratable acidity values were reported between 4.58-4.65 and 0.686-0.803%, respectively while the average total dry matter was between 10.8 and 11.7%. For kefir drink prepared by different percentages of kefir grains and storage days, Irgioyan et al. [18] reported that the average ranges for the pH, fat and dry matter contents of kefir drinks were 4.4-4.7, 3.23-3.60% and 11.3-11.7%, respectively. Ünal et al. [12] reported that the ranges for the dry matter, pH and acidity values of commercial kefir samples were 8.71-10.68%, 4.26-4.37, and 0.67-0.88%, respectively. Moreover, the fat content of commercial kefir drinks was reported between 0.55 and 2.90%.

Eryilmaz [5] determined the chemical and mineral composition, and antioxidant properties of kefir drink produced with different milk samples and kefir cultures. In this study, the dry matter content (11.1-12.0%), pH (4.64-5.88) and titratable acidity (0.42-0.86%) values of kefir samples were found similar for different milk samples (p>0.05). Studying the influences of fermentation process variations on kefir quality, Kök-Taş et al. [11] reported the pH and lactic acid (%) values in the ranges of 4.29-4.53 and 0.81-0.95%, respectively. Also, the protein and total solids contents (%) of kefir drinks were in the ranges of 3.09-3.48% and 7.81-8.21% during different days of cold storage. Results for the chemical properties of commercial kefir drinks in this present study were in good agreement with those reported in the literature.

Color parameters (L*, a* and b*) of various commercial kefir drinks are presented in Table 2.
The L*, a* and b* color values of commercial kefir samples were in the intervals of 84.21 and 86.68, -2.87 and -2.40, 4.80 and 6.03, respectively. The highest L* color value was obtained for brand A (p<0.05), while the lowest L* color value was obtained for brand F (p<0.05). Doğan [19] determined the Hunter L color values of kefir with honey as between 63.10 and 73.18. Also, Hunter a and b color values were determined between -1.50 to -0.92 and 3.77 to 9.35, respectively. Atalar and Derişoğlu [20] determined the Hunter L, a and b color values of kefir powders as between 84.04 and 90.6, -2.94 and -1.09, 8.62 and 14.64, respectively. Goncu et al. [21] reported that the Hunter L, a and b color values of kefir samples enriched with apple and lemon fiber changed between 67.23 and 78.87, -1.55 and 1.46, and 9.17 and 11.62, respectively.

### Rheological Properties

The apparent viscosity, consistency coefficient and flow behavior index values of commercial kefir samples are presented in Table 3 for different brands at three temperatures.

| Sample code | Temperature (°C) | Apparent viscosity at 111.6 s⁻¹ (mPa.s) | Flow behavior index (n) | Consistency coefficient (K, mPa.s) | Coefficient of determination (R²) |
|-------------|-----------------|----------------------------------------|-------------------------|------------------------------------|----------------------------------|
| A           | 4               | 1.24±0.08DE                           | 0.44±0.04A              | 12.12±2.07HF                       | 0.996                            |
|             | 10              | 1.76±0.03A                            | 0.34±0.02BC             | 31.23±3.06AB                       | 0.996                            |
|             | 25              | 0.48±0.09G                            | 0.18±0.05F              | 28.44±8.54AD                       | 0.963                            |
| B           | 4               | 1.43±0.12BC                           | 0.30±0.02CD             | 32.84±4.16A                        | 0.994                            |
|             | 10              | 1.76±0.03A                            | 0.34±0.02BC             | 31.23±3.06AB                       | 0.996                            |
|             | 25              | 0.91±0.11F                            | 0.30±0.04CD             | 20.22±3.41AD                       | 0.995                            |
| C           | 4               | 1.46±0.05BC                           | 0.32±0.03BC             | 28.58±3.83AD                       | 0.986                            |
|             | 10              | 1.43±0.09BC                           | 0.31±0.04CD             | 30.37±7.37AB                       | 0.994                            |
|             | 25              | 0.52±0.14G                            | 0.23±0.07EF             | 20.80±5.64CG                       | 0.973                            |
| D           | 4               | 1.29±0.06CE                           | 0.34±0.02BC             | 21.83±2.24DG                       | 0.997                            |
|             | 10              | 1.18±0.08E                            | 0.32±0.01BC             | 22.86±2.62CG                       | 0.993                            |
|             | 25              | 1.55±0.09FD                           | 0.31±0.06CD             | 13.66±4.26EF                       | 0.979                            |
| E           | 4               | 1.59±0.09AB                           | 0.34±0.03BC             | 27.37±4.92DE                       | 0.996                            |
|             | 10              | 1.21±0.35DE                           | 0.32±0.06BC             | 23.19±4.72DG                       | 0.993                            |
|             | 25              | 0.58±0.11G                            | 0.22±0.05EF             | 24.80±5.51IL                       | 0.983                            |
| F           | 4               | 1.59±0.06AE                           | 0.34±0.02BC             | 27.65±2.66AC                       | 0.997                            |
|             | 10              | 1.35±0.08CD                           | 0.31±0.03CD             | 29.66±8.28IC                       | 0.996                            |
|             | 25              | 0.57±0.06A                            | 0.26±0.03DE             | 16.54±1.97IG                       | 0.981                            |

Table 3 demonstrated that the Power Law model was appropriate to interpret the rheological characteristics of kefir drinks with high R² values ranging from 0.963 to 0.997 (Table 3). This model was previously used to determine the rheological flow properties of fermented milk products as plain yoghurt [22], concentrated milk [9], acidified milk drinks [23], yoghurt drink [24] and kefir [11, 18, 25]. Apparent viscosity values of commercial kefir samples ranged from 0.48 to 1.76 mPa.s. Apparent viscosity values of commercial kefir samples were higher at 4°C than those at either 10 or 25°C. Results showed that the kefir samples of brand B, E and F were more viscous at 4°C. Also, brand B had a higher apparent viscosity value, in general, at 4 and 25°C than the others (p<0.05). Flow behavior index values (n) changed from 0.18 to 0.44, indicating a typical pseudoplastic behavior. The significantly highest n value was observed for brand A at 4°C (p<0.05). The flow behavior type of commercial kefir samples was non-Newtonian pseudoplastic with respect to Power Law indices (n<1) for all samples (p<0.05).

Similar rheological analysis results were obtained for kefir drinks in the literature studies. Goncu et al. [21] reported the viscosity of kefir samples enriched with apple and lemon fiber in the range of 0.67 and 9.73
mPa.s. Studying the rheological properties of kefir samples produced by mare’s milk and its mixtures with goat and sheep milk, Cais-Sokolińska et al. [26] determined the viscosity values of samples between 3.09-4.45 mPa.s. In addition, Garrote et al. [27] reported the apparent viscosity of different kefir samples prepared by different kefir grains between 7.5 and 15.4 mPa.s.

In a study [17], power law model was found appropriate for defining the rheological behavior of kefir drinks with flower and pine honey. In this study, kefir beverages exhibited a non-Newtonian behavior, where the flow behavior indexes (n) changed from 0.37 to 0.47. Similar results for yoghurt drinks were reported in a study by Gursoy et al. [24] where the influence of ultrasound power on the rheological properties of yoghurt drinks was investigated. According to our results, power law model was suitable to explain the rheological characteristics of kefir samples, which showed a non-Newtonian type flow behavior. Studying the rheological characteristics of a kefir drink at various temperature levels, İcier et al. [13] reported a non-Newtonian pseudoplastic flow behavior with the flow behavior index values of 0.410, 0.426 and 0.351 at 10, 20 and 30°C, respectively. Ünal et al. [12] reported the flow behavior index values of commercial kefir samples ranging between 0.26 and 0.45.

The activation energies of commercial kefir samples were determined by plotting ln(η) versus (1/T) as shown in Figure 1.

![Arrhenius plot of apparent viscosity values of commercial kefir drinks](image)

**Figure 1.** Arrhenius plot of apparent viscosity values of commercial kefir drinks

Activation energy values of kefir samples were determined by the Equation 2 using Figure 1 and changed between 21.51 and 36.18 (Table 4). Results indicated that the viscosity of brand C is relatively more sensitive to temperature changes in comparison to kefir drinks of other brands. Studying the effect of heating from 22 to 70°C on the rheological properties of Cheddar, Colby, whole milk Mozzarella, low-moisture part-skim Mozzarella, Parmesan, soft goat, and Queso Fresco cheeses, Tunick [28] found the activation energy values of cheeses between 30.4 and 182.5 kJ/mol. In a study determining the temperature dependency of apparent viscosity of oat milk, activation energy values were reported between 07.43 and 303.64 kJ/mol [29]. Abu-Jdayil et al. [30] determined the activation energy of concentrated yogurt (Labneh) between 20.92 and 21.88 kJ/mol.

| Sample code | Activation Energy (kJ/mol) |
|-------------|---------------------------|
| A           | 32.87                     |
| B           | 21.51                     |
| C           | 36.18                     |
| D           | 31.25                     |
| E           | 33.25                     |
| F           | 34.77                     |

Table 4. Activation energies of commercial kefir drinks

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Correlation among Physicochemical and Rheological Properties

The Pearson correlation coefficients for the physicochemical properties of commercial kefir samples by their rheological properties are shown in Table 5. According to Table 5, the correlation of physicochemical properties of kefir samples with apparent viscosity values were statistically insignificant (p>0.05); however, correlations of fat (%), pH and b* (yellowness) color value with K and n values of commercial kefir samples were found statistically significant (p<0.05). Accordingly, as fat (%) and pH values of kefir samples increased, their K values decreased while their n values increased (p<0.05).

|                      | Dry matter | Protein | Fat content (Dry matter basis) | Fat (%) | pH | Total titratable acidity | L* | a* | b* |
|----------------------|------------|---------|--------------------------------|---------|----|--------------------------|----|----|----|
| Apparent viscosity   | -0.2154    | 0.1601  | -0.1185                        | -0.3643 | -0.4023 | -0.3530                | -0.3189 | 0.2895 | -0.3883 |
|                      | (0.5014)   | (0.6191) | (0.7138)                       | (0.2444) | (0.1948) | (0.2604)               | (0.3123) | (0.3614) | (0.1998) |
| K                    | -0.4868    | 0.0990  | -0.1198                        | -0.5962 | -0.7170 | -0.0025                | -0.4103 | 0.0815 | -0.6225 |
|                      | (0.1085)   | (0.7596) | (0.7107)                       | (0.0408)** | (0.0087) | (0.9938)               | (0.1853) | (0.8011) | (0.0306) |
| n                    | 0.5510     | -0.1210 | 0.1079                         | 0.62554 | 0.8574 | -0.2608                | 0.4389 | 0.1225 | 0.6194 |
|                      | (0.0634)   | (0.7081) | (0.7385)                       | (0.0296) | (0.0004) | (0.4129)               | (0.1534) | (0.7045) | (0.0317) |

** Bold values show statistically significant correlations (p<0.05)

CONCLUSION

Kefir is a popular dairy product with its functional properties and probiotic content. In the present study, physicochemical and rheological properties of the kefir drinks containing the most popular and dominant brands of the Turkish kefir market were determined. Results showed that significant differences in physicochemical and rheological properties were present among the commercial kefir samples of different brands, indicating the compositional variations of raw materials and kefir cultures used in their production. Additionally, rheological properties were influenced by temperature, and Power Law model could be used to define their flow behavior, which was of a pseudoplastic type over the temperature range studied. Fat (%), pH and CIE b* color values of commercial kefir samples were found to be inversely correlated with consistency coefficient values (K), and these values were positively correlated with the flow behavior index values of commercial kefir drinks (n). This study may promote an important understanding of the physicochemical properties and rheological behavior of the commercial kefir beverages marketed in Turkey.

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