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Original Research

Technological, microbiological and sensory quality attributes of goat's milk kefir: comparison between plain and flavored types

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Abstract:

Introduction: Kefir is a popular probiotic drink that strengthens the immune system, reduces lactose intolerance and lowers blood cholesterol levels. It can be made from various milk sources, including goat's milk, which has superior nutritional qualities such as higher protein content and better digestibility compared to cow's milk. The objectives of this study were: firstly, to optimize the production parameters for goat's milk kefir, focusing on the incubation time, temperature and grain/milk ratio to achieve standard technological and microbiological qualities of kefir; and secondly, to monitor the changes in the physicochemical, textural and microbiological properties of kefir during shelf life. Two types of kefir (plain and flavored) were assessed and also compared in terms of consumer preference.

Methods: The optimization of the incubation time, temperature and grain/milk ratio was carried out by using a Box-Behnken design and response surface to analyse the pH, acidity, syneresis, mesophilic and lactic acid bacteria concentrations, firmness, consistency, cohesiveness and viscosity index of plain kefir (control). After optimizing the production process, the physicochemical, textural and microbiological quality properties of plain and lemon-flavored kefir were assessed over time during a two-week storage at 4 ºC. The treatments were compared by linear modelling. The sensory attributes of appearance, odor, taste, sourness, smoothness and acceptability were assessed by a consumer panel and statistically analyzed on variance to determine differences between the two kefir types.

Results: The optimization experiment revealed significant effects ($p < 0.05$) of three factors on quality properties of kefir. The acidity was maximized at a 0.66% grain/milk ratio, 25.3 °C and 22.8 h, with incubation temperature having the greatest impact. Specific conditions optimized technological properties, particularly firmness (> 5 g) and viscosity index (< 1 g.s). Syneresis varied greatly with temperature; the highest values were found at lower temperatures (15-20 °C) and shorter incubation times (16 h). However, lower temperatures or longer incubation times could produce the desired acidity and texture, but only at higher grain/milk ratios (>1.0%). In the second phase of the experiment, the flavored kefir maintained a lower pH (4.00), which was beneficial for pathogen inhibition, and exhibited less pronounced proteolysis, contributing to better textural stability. While both kefir types reached similar firmness and cohesiveness, the flavored kefir showed a higher viscosity index and consistency, which had a positive effect on mouthfeel and consumer preference. Microbiologically, the control kefir presented slightly higher lactic acid bacteria and mesophilic populations (7.7 log CFU/mL and 7.6 log CFU/mL, respectively) than the flavored kefir (7.4 log CFU/mL and 7.2 log CFU/mL, respectively). The sensory evaluation revealed a clear preference among panelists for lemon-flavored kefir in almost all sensory attributes.

Conclusion: This comprehensive study offers crucial insights into the production of goat's milk kefir by elucidating the effects of key processing variables on product quality. It provides valuable information on the quality

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characteristics and storage stability of both the natural and flavored variants of kefir and enhances our understanding of quality changes over time. These findings not only contribute to optimizing production parameters for high-quality goat's milk kefir, but also lay a solid starting point for future research in this field.

Keywords: Kefir grains, Box-Behnken, Linear modeling, Response surface, Acidity, Hardness, Viscosity, Lactic acid bacteria, Cold storage

1. Introduction

Kefir, one of the most popular probiotic drinks, is a notable example of a beneficial fermented food known and recognized globally for its nutritional value. This thick, creamy drink with a slightly fizzy, sour taste and low alcohol content has been demonstrated to strengthen the immune system, reduce lactose intolerance and lower blood cholesterol levels [1]. Recent advances in goat's milk kefir highlight enhanced understanding of fermentation and probiotic benefits such as blood sugar control and anti-inflammation. The global market for goat's milk products is booming and is expected to reach USD 20.75 billion by 2029, driven by demand for cheese, infant formula and nutritional benefits. However, the kefir industry faces production challenges such as safety regulations and market competition, while the kefir market is expected to reach USD 3.68 billion by 2033 [2]. Published in 2023, a study using metagenomic analysis revealed that goat's milk kefir has a higher prevalence of Firmicutes, including the genera Lactobacillus, Streptococcus and Staphylococcus, compared to cow's milk kefir. However, its yeast population is limited, with only 2% belonging to the Saccharomycetaceae family, while the majority remain unclassified. This microbial composition underscores the unique fermentation dynamics and probiotic potential of goat's milk kefir [3].

Kefir is produced by fermenting goat's, cow's, camel's or sheep's milk with kefir grains, which are a symbiotic culture of yeasts and bacteria encased in a matrix of polysaccharides. These grains play a crucial role in creating kefir's distinctive characteristics [1]. A strong symbiotic homeostasis system exists in goat's milk kefir [4], which stands out by its unique fat composition: smaller fat globules, a higher proportion of short and medium chain fatty acids, and a softer curd structure. These characteristics contribute to enhanced digestibility compared to other types of milk. Consequently, goat's milk supports a more efficient lipid metabolism and offers potential health benefits [5]. It is important to note that the physicochemical and microbial quality of kefir made from fermented milk is influenced by multiple factors, including milk type, grain/milk ratio, fermentation conditions and storage temperature. Maintaining hygienic and sensory standards is crucial in kefir production. In this context, the present study aims to investigate the changes in technological and microbiological properties of plain goat's milk kefir over time. The objectives of this study were: (1) to optimize the production parameters for goat's milk kefir, focusing on the incubation time, temperature and grain/milk ratio to achieve standard technological and microbiological qualities of kefir; and (2) to assess the deterioration pattern of kefir along its shelf life by monitoring the changes in the physicochemical, textural and microbiological properties of two types of kefir (plain and flavored) during refrigerated storage. Finally, the two types of goat's milk kefir were compared in terms of consumer preference through a sensory evaluation.

2. Methodology

The present work was divided into two phases: the first phase was to optimize the conditions (grain/milk ratio, incubation time and incubation temperature) for the production of kefir with standard technological quality. The fermentation parameters, including the type of kefir culture (natural grains or starter cultures), inoculation ratio, temperature and duration, significantly affect the microbial composition, chemical properties and sensory qualities of kefir [6]. The selection of grains/milk ratio, incubation time and temperature for kefir optimization is supported by extensive research demonstrating that they have a significant impact on the product's technological and microbiological qualities. A higher grains/milk ratio generally results in faster fermentation and more acidic kefir, while a lower ratio allows for slower fermentation and potentially different flavor profiles [7]. The incubation time is a critical parameter in kefir production as it directly influences the extent of fermentation and affects the final pH value, the microbial counts and the sensory attributes of the product. Longer fermentation times lead to increased acidity and higher counts of lactic acid bacteria and yeasts. The optimal incubation time can vary widely, typically ranging from 10 to 48 hours, depending on the desired product characteristics such as the target acidity level, the number of probiotic bacteria, the flavor profile and the texture preferences [8].

Higher temperatures (33-35°C) promote yeast growth in kefir, while the incubation temperature affects the production of flavor and aroma compounds. These compounds include acetaldehyde, diacetyl, acetoin and ethanol, which contribute to the characteristic taste and smell of kefir [9].

In the second phase, the changes in the physicochemical, microbiological and textural quality attributes of two types

of kefir (plain and lemon-flavored) produced under the optimized conditions were evaluated during cold storage. Furthermore, these two types of kefir were compared in terms of sensory attributes, which were evaluated by a panel of potential consumers. Section 2.1 elaborates the kefir manufacture process as well as each of the physicochemical, microbiological, textural and sensory analysis carried out on kefir samples. Subsequently, the first and second phases of the work are described in sections 2.2 and 2.3, respectively.

2.1 Description of analysis

Kefir manufacture

The kefir grains used in this study originate from Tunisia. They were already in stock in the freezer of the CIMO laboratory, as they had previously been used for the production of camel milk kefir. To produce kefir, the kefir grains were activated in pasteurized goat's milk (President, Lactalis, Portugal): lipids 1.5 g; carbohydrates 4.5 g; proteins 3.3 g; salt 0.2 g; calcium 120 mg in 100 ml) for 24-48 hours in an incubator (25°C) to initiate microbial activity. Once activated, the grains were added to pasteurized goat's milk in a weight ratio of 5-10% to create optimal conditions for fermentation. The mixture was fermented in an incubator at 20-25°C for 12-24 hours. During this time, the texture, flavor and acidity changed. Following fermentation (in a 1 liter glass flask with 75 cm headspace), the kefir grains were carefully separated from the milk by gravity filtration using high quality filter paper (size 240 mm diameter) to ensure a smooth and uniform final product. The filtered kefir was then transferred to sterile containers and stored at 4 °C, a temperature chosen to preserve quality and maintain microbiological integrity throughout the shelf life.

Physicochemical analysis

To assess the physicochemical quality of each kefir treatment, we performed a comprehensive set of analyses, including pH, titratable acidity, syneresis and proteolytic activity. These analyses were carried out in duplicate.

pH: pH was determined using a FiveGo pH meter F2 coupled with a LE438 IP67 probe (Mettler-Toledo, Greifensee, Switzerland).

Acidity: Acidity was measured through titration with sodium hydroxide (NaOH) using phenolphthalein as an indicator. For this purpose, a 10 mL kefir sample was prepared in a beaker, two drops of phenolphthalein were added and titrated with 1/9 N NaOH (Dornic solution) until a stable pink color appeared, signifying the endpoint. The titrable acidity (TA), expressed in Dornic degrees (°D), was calculated by multiplying the volume of NaOH used (in mL) by 10, where 1 °D corresponds to 0.01 grams of lactic acid per liter of kefir [10]. The procedure was repeated three times per sample with the average value recorded as the final result.

$$TA\ (°\mathbf{D}) = \mathbf{V}_i * 10$$

Syneresis: Following the method described by Schmidt and Bouma, we developed an in-house protocol to measure syneresis) [11]; the procedure involves separating and quantifying the amount of free whey to assess whey separation, an important quality indicator in fermented dairy products. A 20 g kefir sample was first centrifuged at 3000 rpm for 20 minutes at 4°C to separate the solid and liquid phases in order to monitor the water-holding capacity of kefir. This was followed by filtration to isolate the liquid whey. The syneresis was calculated as a percentage of the free whey according to the following formula:

$$\text{Free whey or synereis (\%)} = PFSW * 100 / ISW$$

where PFSW is the sample weight after filtration and ISW is the weight of initial sample in grams.

Proteolytic activity: Proteolysis was quantified using an in-house protocol by first adjusting the sample's pH to 4.6 with 1 M HCl to precipitate the casein, followed by centrifugation of a 10 mL of sample at 3000 rpm for 20 minutes at 4°C to separate the whey, which was then filtered for analysis (240 mm diameter filter). To quantify the proteolytic activity in kefir, the following procedure was followed: First, 1 mL of whey was diluted in 100 mL of distilled water and the absorbance was measured at 280 nm, with distilled water serving as a blank standard. The remaining whey sample was then subjected to heat treatment by boiling in a water bath for 10 minutes. It was then centrifuged at 3000 rpm for 20 minutes at 4°C and the resulting liquid was filtered (240 mm filter diameter). Finally, the filtered, boiled whey was diluted by adding 1 mL of the sample to 10 mL of distilled water, and the absorbance was measured again at 280 nm using distilled water as a blank. With this method, the proteolytic activity in kefir can be evaluated by comparing the absorbance before and after heat treatment.

$$\textit{Proteolytic activity (no units)} = \textit{Filtrate absorbance before boiling - Filtrate absorbance after boiling}$$

Textural analysis

We evaluated the texture of kefir using a Texturometer equipment (TA. XT Plus) calibrated with a 5 kg load cell. A flat cylindrical probe (P/36R, 36 mm diameter) was used for texture profile analysis (TPA). The kefir sample (80 mL) was placed in a jar and positioned under the probe for measurement at room temperature. The test procedure included several steps: switching on the machine, starting the Exponent software, calibrating the height to 80 mm, setting the probe position to 49 mm and

configuring the settings for a 20 mm penetration distance (two compression cycles at a speed of 2 mm/s).

Texture Exponent TPA32 software was used and quantified key parameters: firmness: resistance to deformation (g); viscosity index: flow resistance indicating thickness (g.s); cohesiveness: ability to retain structure under compression (g); and consistency: uniformity and stability of texture (g.s). This comprehensive analysis provided insights into kefir's structural integrity, thickness, texture retention and overall uniformity, essential factors in evaluating product quality.

Microbiological analysis

The following determinations were carried out following decimal serial dilutions, starting from a first dilution of 1 ml kefir in 9 ml buffered peptone water. The number of replicates per kefir sample was set to two.

Lactic acid bacteria (LAB): For the enumeration of LAB in the kefir samples, we employed MRS (de Man, Rogosa and Sharpe) agar as culture medium with 1 mL of Tween 80 (Frilabo, 80031, Portugal) added per liter of medium to enhance LAB growth. Inoculation was performed in a double layer, where 1 mL of homogenate was placed in a Petri dish. A base layer of MRS agar is poured over it, swirled, and allowed to solidify. Then, a second agar layer was poured on top and left to dry, creating a layered environment for LAB growth, incubating anaerobically at 30°C/48h in a candle jar.

Total mesophiles (MES): PCA (Plate Count Agar) was employed for enumerating mesophilic bacteria, which provided a non-selective environment for a broad spectrum of mesophilic organisms. Incubation was performed aerobically at 37°C for 48h following the standard protocol NF V08-011, 1998. This bacterial count is an indicator of overall food quality and hygiene during processing, storage and handling. It helps detect spoilage early, allowing for corrective actions and extended shelf life. Although safety was not measured directly, high bacterial counts can prompt further safety testing. Tracking MES over time helps assess product stability and optimize storage practices.

Yeast and molds: Such counts were performed using 3M Petrifilm™ Yeast and Mold Count Plates, which contain a colored indicator for easy differentiation between yeast and mold colonies. This microbiological analysis involved incubating the samples aerobically for 3 to 5 days at 25°C, using dilutions ranging from 10^0 to 10^{-2}. The procedure involved placing 1 mL of the diluted sample in the center of the bottom film of the Petrifilm, which was then spread evenly using the 3M™ Petrifilm™ Yeast and Mold Spreader, applying light pressure without twisting or sliding the spreader. After incubation, colonies were counted using a standard electronic colony counter, with yeast colonies appearing as small, defined blue-green colonies and mold colonies appearing as larger, more diffuse colonies with variable colors.

Microbial concentrations were all calculated in log base 10 CFU/ml from plates containing colonies in the range of 20-200.

Sensory analysis

To quantify the sensory experiences and compare two kefir samples (control and flavored), prepared one day before the test . A non-structured 9-point scale was used for evaluation. Participants (IPB and CIMO students and researchers with an average age of 30 were randomly selected) assessed appearance, odor, taste, sourness, smoothness and overall acceptance. A response sheet was designed with separate rows for each three-digit coded sample, allowing 45 participants to mark their ratings. After tasting, the distances from the left edge of the scale to each mark were measured in centimeters for precise data collection. These measurements were then organized in a data table for subsequent statistical analysis.

2.2 Optimization of conditions for kefir production

Fourteen kefir treatments were conducted using a Box-Behnken design with three central points, incorporating three factors: grain/milk ratios (0.5%, 1%, and 1.5%), incubation time (16, 20, and 24 hours), and incubation temperature (15°C, 20°C, and 25°C) (Table 1). Each factor, tested at three levels: low (-1), medium (0), and high (+1), resulted in a 14-treatment experimental matrix. This experimental design allowed the evaluation of the impact of each variable and their interactions on each of the kefir's quality properties, in order to determine the optimal conditions for kefir production. We assessed the effectiveness of these conditions through a comprehensive analysis of kefir samples stored at 4°C. We examined key properties including pH, acidity (% lactic acid), syneresis (%), concentration of mesophilic and lactic acid bacteria, firmness, consistency, cohesiveness and viscosity index to determine the most favorable incubation parameters for producing kefir of standardized quality.

Data analysis was performed by fitting a response surface model to each of the quality attributes using the rsm package of R software. Analyses of variance (ANOVA) were performed at $\alpha=0.05$ to identify the significant linear, interactive and quadratic terms. This approach allowed us to optimize the experimental conditions for kefir incubation covering all quality properties.

Table 1. Box-Benhken experimental design

Number	Ratio	Time	Temperature
1	0.5	20	15
2	1.0	16	25
3	1.5	20	25
4	0.5	16	20
5	1.5	20	15
6	1.0	24	15
7	1.5	16	20
8	1.0	20	20
9	1.5	24	20
10	0.5	20	25
11	1.0	20	20
12	1.0	24	25
13	1.0	16	15
14	0.5	24	20

Factors levels– kefir: 0.5, 1.0 and 1.5%– Time: 16, 20 and 24 hours– Temp: 15, 20, 25 ºC

2.3 Changes in physicochemical, textural and microbiological attributes

In the second phase of our study, we conducted a comprehensive comparison between two kefir varieties: a control sample and a lemon-flavored variant enhanced with lemon extract. We added lemon extract (20 ml bottle of Vahiné, Spain) at a concentration of 1% to the milk and grains prior to fermentation. This addition was intended to enhance the flavor profile and potentially influence the fermentation process, allowing the lemon extract to blend with the milk and grains before fermentation began. The 1% dosage was carefully measured to achieve the desired effect without overwhelming the final product.

While the first phase of this study enables to set the conditions (incubation temperature, time and kefir grains/milk ratio), the second phase of the study focused on performing a wide range of quality attributes essential for assessing product integrity, consumer appeal and shelf stability of kefir, including physicochemical attributes (pH, acidity, syneresis and proteolysis), textural features (firmness, viscosity index and consistency), microbiological profiles (focusing on the number of lactic acid bacteria and mesophiles) during a 13-day storage at 4 ºC. This comprehensive approach provided valuable insights into the impact of lemon flavoring on kefir's quality and stability across multiple dimensions and offered a thorough understanding of the product's characteristics and shelf-life under optimal storage conditions. Furthermore, the sensory analysis of the two kefir types was carried out on samples stored at 4 ºC for a maximum of two days.

Data analyses by linear modelling was carried out to determine the differences between the kefir types and the effects of storage time on each of the quality attributes assessed. In addition, scatter plots were created to illustrate the evolution of the quality parameters over time by kefir type. For the sensory evaluation data, analyses of variance were conducted on each of the sensory attributes to determine any significant differences between kefir types. A radar graph was also created. All of these statistical analyses were undertaken using R software.

3. Results and discussion

The quality of kefir is significantly influenced by the ratio of kefir grains/milk and the fermentation conditions, namely incubation time and temperature. In the first phase of our study, through response surface analysis we identified the parameters for producing kefir of standard physicochemical, textural and microbiological qualities. In the second phase of the study, we used these established baseline conditions to monitor the changes of the quality parameters of two types of kefir along shelf life.

3.1 Results of the optimization of conditions for kefir production

Physicochemical properties

The investigation showed that the examined factors significantly impacted kefir's quality characteristics, with each primary factor exerting a measurable and meaningful effect on the product's properties (Table 2). Two-way interactions between ratio and time, as well as time and temperature, notably impacted pH and acidity ($p < 0.05$). Additionally, the quadratic effects of time and temperature exhibited a significant influence on the majority of physicochemical attributes in kefir production. However, these quadratic terms did not demonstrate a notable impact on syneresis.

Among the treatments, the pH of kefir ranged between 3.65 (grains/milk ratio: 1%, 16 h, 15 ºC) and 6.23 (grains/milk ratio: 1%, 24 h, 25 ºC). The response surface analysis

of kefir pH, influenced by temperature and kefir grains/milk ratio, highlighted their impact on the acidity of final products (Figure 1). The study found that pH peaks under specific conditions (0.48% ratio, 25.3ºC, 24.2 hours), with the grain ratio being the key factor affecting pH levels. In line with these findings, Dewi et al. (2020) found that the pH of goat's milk kefir is highest at low kefir grain concentrations and without storage, and lowest at high concentrations after 21 days, indicating that both factors significantly affect pH [5].

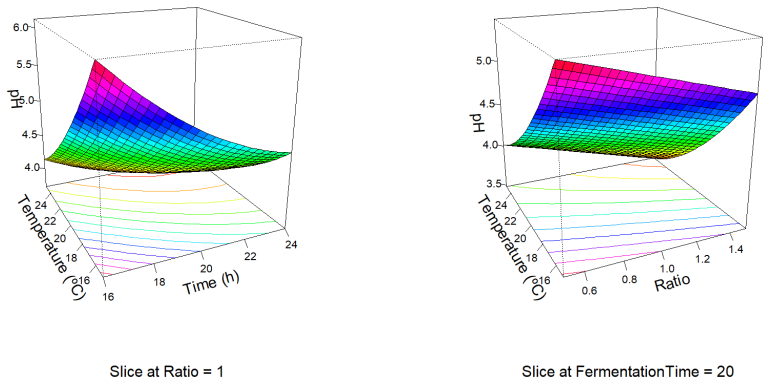

Figure 1. Response surface analysis of kefir pH as a function of temperature incubation time and grains/milk ratio, illustrating optimal conditions for pH

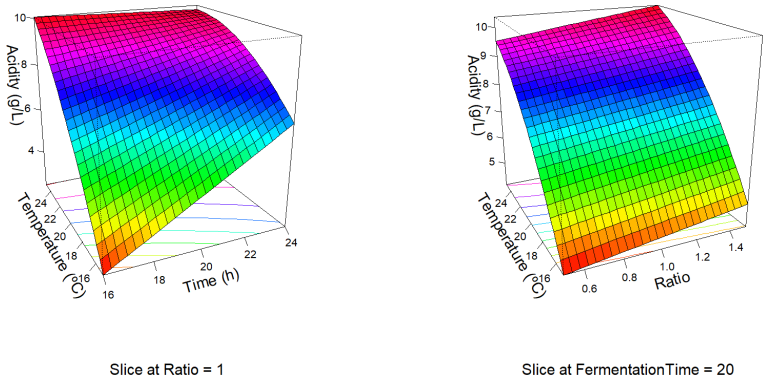

Figure 2. Response surface analysis of kefir's acidity (expressed as lactic acid in g/L) as a function of temperature, time and grains/milk ratio, illustrating zone of optimal conditions for acidity

Among the treatments, the acidity of kefir ranged between 2.38 g/L (grains/milk ratio: 1%, 16 h, 15 ºC) and 10.40 g/L (grains/milk ratio: 1.5%, 20 h, 25 ºC). By response surface analysis, kefir acidity was found to peak at a ratio of 0.66% grains/milk, a temperature of 25.3ºC, and an incubation time of 22.8 hours (Table 2). Temperature has the greatest influence on acidity, followed by incubation time, while the grains/milk ratio has the least influence (Figure 2). Aligned with these findings, Putri, Setiani, and Warya found that optimizing incubation time and temperature is crucial for maximizing lactic acid production in kefir fermentation. Their study showed that longer fermentation and higher temperatures resulted in higher concentrations of organic acids [12]. The right level of acidity contributes to the characteristic tangy flavor of kefir, which is essential for its unique appeal. Consumers generally prefer kefir with a slightly sour taste that is not too overpowering [13].

Syneresis was the only physicochemical property that was affected only by the first-order (linear) terms (Table 2). At an incubation temperature of 25°C, the syneresis of goat's milk kefir ranged from 38.92% to 61.47%. In contrast, at the minimum temperature of 15°C, the syneresis of kefir reached notably high levels,

ranging from 93.97% to 95.54% (Figure 3). These high syneresis values highlighted the substantial effect of low temperature on the moisture retention properties of kefir.

Textural properties

The research revealed a significant interplay between incubation time and temperature, which substantially influenced kefir's textural attributes such as firmness, consistency and cohesiveness. Additionally, the quadratic terms for ratio and temperature demonstrated notable impacts on the majority of textural properties (Table 2). Interestingly, the viscosity index stood out as an exception as it was not significantly affected by these quadratic terms. These results underscored the sensitivity of kefir's microstructure to thermal exposure, which impacts protein gel network formation and moisture retention.

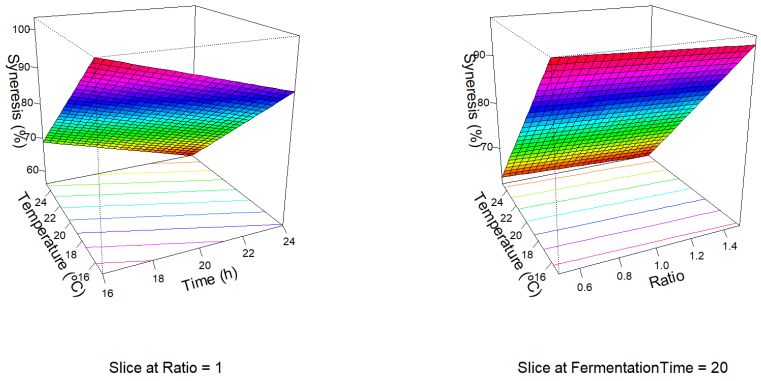

Figure 3. Response surface analysis of kefir's syneresis (%) as a function of temperature, time and grains/milk ratio

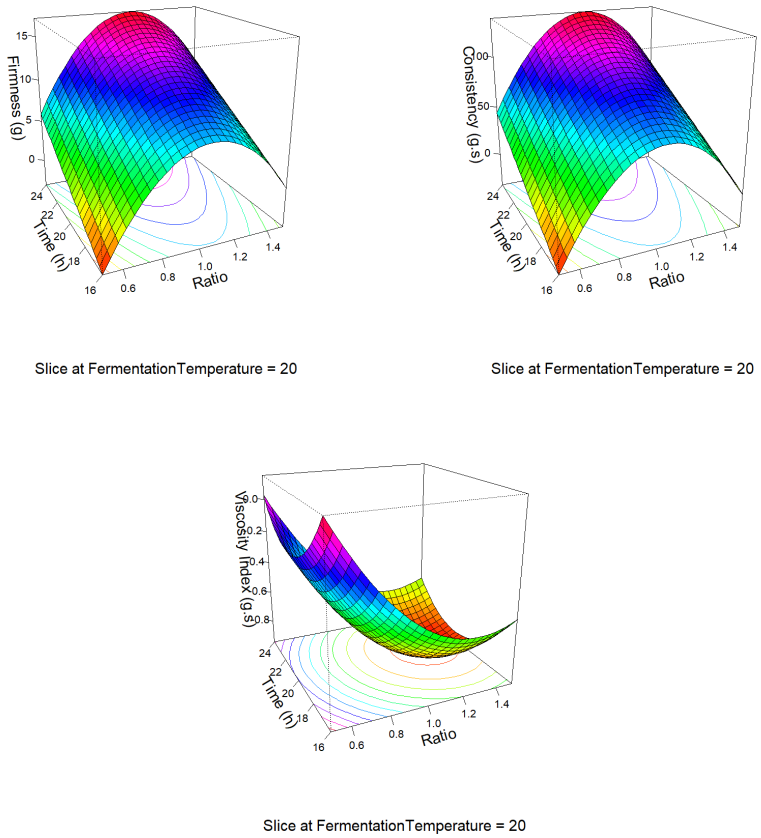

Figure 4. Response surface analysis of kefir's firmness (g), consistency (g.s) and viscosity index (g.s) as a function of incubation time and grains/milk ratio, illustrating optimal conditions for textural parameters.

Table 2. Analysis of variance of the response surface models fitted to all the quality attributes measured in goat's milk plain kefir
FO: Factorial Optimization, TWI: Two-Way Interaction, Pr: Probability, F: F-statistic, R^2adj Adjusted R-squared, P value: Probability value

	pH		Acidity		Syneresis		Firmness		Consistency		Viscosity Index		LAB		MES	
	Mean Sq	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**	**Mean Sq**	**Pr(>F)**
FO	4.119	<0.0001	45.380	<0.0001	1785.58	<0.0001	277.90	0.005	21613	0.006	1.154	0.004	6.960	<0.0001	5.452	<0.0001
TWI	-	-	-	-	-	-	-	-	-	0.025	-	-	-	-	-	-
Ratio. Time	0.905	<0.0001	5.445	0.003	-	-	-	-	-	-	-	-	-	-	-	-
Time Temp	0.543	0.003	10.31	0.0002	-	-	592.20	0.002	44104	0.002	-	-	-	-	-	-
Ratio.temp	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Ratio²	-	-	-	-	-	-	679.26	0.001	50182	0.001	-	-	4.368	0.011	-	-
Time²	0.258	0.034	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Temp²	0.295	0.024	4.122	0.009	-	-	368.17	0.011	23690	0.018	-	-	2.588	0.049	1.787	0.052
Residuals	0.05	-	0.495	-	61.53	-	-	-	3573	-	0.2	-	0.573	-	0.428	-
Pure error	0.003	-	0.111	-	44.62	-	-	-	188	-	0.13	-	0.227	-	0.222	-
Stationary point (T in °C, Time in h)	R=0.48 T=25.3 Time =24.2		R=0.66 T=25.3 Time =22.8		R=1.673 T=8.9 Time =35.7		R=1.07 T=19.9 Time=17.4		R=1.07 T=19.9 Time=17.4		R=1.28 T=18.5 Time=20.4		R=0.61 T=21.4 Time=23.2		R=0.33 T=21.9 Time=21.2	
R^2adj P-value	0.913 <0.0001		0.92 <0.0001		0.76 <0.0001		0.628 <0.0001		0.626 <0.0001		0.5202 0.001		0.617 <0.0001		0.587 <0.0001	

Table 3. Analyses of variance assessing the impact of storage time and type of kefir on the evolution of all quality attributes measured in goat's milk plain and flavored kefir stored at 4 ºC

	pH				**Acidity**				**Syneresis**				**Proteolysis**			
Source of variation	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value
Intercept	1	35	60175	<.0001	1	31	59813	<.0001	1	17	3874.8	<.0001	1	16	150.951	<.0001
Kefir type	1	35	0	0.9274	1	31	88	<.0001	1	17	0.4	0.5435	1	16	14.034	0.0018
Day	1	35	0	0.7618	1	31	11	0.0023	1	17	27.0	0.0001	1	16	2.400	0.1409
Type : Day	1	35	1	0.3572	1	31	14	0.0008	1	17	0.0	0.8583	1	16	5.230	0.0362
	Firmness				**Consistency**				**Viscosity index**				**Cohesiveness**			
Source of variation	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value
Intercept	1	31	8080.9	<.0001	1	31	7819.2	<.0001	1	29	4508.1	<.0001	1	31	244.960	<.0001
Kefir type	1	31	1.8	0.11866	1	31	6.7	0.0148	1	29	2.1	0.1534	1	31	0.988	0.3255
Day	1	31	7.7	0.0092	1	31	36.0	<.0001	1	17	6.5	0.0166	1	31	0.451	0.5067
Type : Day	1	31	2.9	0.0973	1	31	35.4	<.0001	1	17	4.1	0.0517	1	31	1.468	0.2348
	LAB				**Mesophiles**				**Yeasts**				**Molds**			
Source of variation	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value	num DF	den DF	F-value	p-value
Intercept	1	31	43132	<.0001	1	31	40852	<.0001	1	35	57.607	<.0001	1	35	30.0520	<.0001
Kefir type	1	31	3	0.091	1	31	3	0.1051	1	35	43.676	<.0001	1	35	0.2945	0.5908
Day	1	31	334	<.0001	1	31	263	<.0001	1	35	31.618	<.0001	1	35	0.5202	0.4755
Type : Day	1	31	25	<.0001	1	31	24	<.0001	1	35	0.183	0.6718	1	35	0.0976	0.7566

num DF: Number of Degrees of Freedom den DF: Denominator Degrees of Freedom

The grain/milk ratio significantly influences kefir's firmness, with extreme ratios (both low and high) reducing firmness (Figure 4). A balanced ratio is crucial for maintaining kefir's structural integrity, which is likely due to the optimal interaction between fermentation agents and milk nutrients. Optimal kefir consistency (>100 g·s) is achieved with a 1% grain/milk ratio and a 24-hour incubation, creating an ideal balance for fermentation and nutrient interaction.

Extreme ratios (<0.6% or >1.4%) significantly decreased kefir's consistency (<50 g·s) (Figure 4) and emphasized the sensitivity of the texture to the grain content. Careful control of incubation parameters is crucial for producing kefir with a stable and desirable texture that meets consumer expectations. Kefir's viscosity index is linearly influenced by incubation time, temperature and the grains/ milk ratio, with higher ratios (>1%) and longer incubation (>20 hours) increasing viscosity. However, to obtain a smoother product, it is recommended to keep the grains/ milk ratio below 1% and to limit the incubation time less than 22 hours (Figure 4). Contrary to Dewi et al.'s findings, higher kefir grain concentrations may not always increase the viscosity of goat's milk kefir, this discrepancy could be due to variations in milk composition and microbial activity between goat's and cow's milk [5].

Consumers prefer kefir with a slightly viscous texture and the right thickness, as opposed to a runny consistency. A higher viscosity creates a thicker, more luxurious texture that many consumers find more appealing [14].

Microbiological properties

The microbiological properties of kefir, notably the populations of lactic acid bacteria (LAB) and mesophiles (MES) populations, are significantly influenced by the incubation temperature, time, and grains/milk ratio (Table 2). The statistical evaluation demonstrated a complex, non-linear relationship between temperature and the microbiological properties of kefir, with conditions for maximizing LAB at 21.9°C for 21.2 hours, and for maximizing MES at 21.4°C for 23.2 hours. The quadratic term of the grains/milk ratio significantly affected only the mesophilic populations, with an optimal ratio of 0.61%. However, both LAB and MES concentrations in kefir were strongly linearly influenced by the grains/milk ratio. The incubation time had a lesser impact, while the temperature affected the LAB and MES levels with specific optimal thresholds (Figure 5). These findings highlighted the importance of controlling temperature and grains/milk ratio in the development of LAB in kefir. After 24 hours of fermentation following inoculation, the population of both lactobacilli and lactococci might reach approximately 10^8 CFU/mL [8]. Although it is important to note that these numbers can vary based on factors such as fermentation time, temperature and the specific kefir grains used. Reaching high numbers of LAB is crucial for achieving a robust probiotic profile and provides valuable insights for developing controlled, reproducible fermentation processes that support stable mesophilic populations in kefir.

Synthesizing our findings, we established that the optimal parameters for kefir production were incubation temperature of 20°C, incubation times in the range of 20-24 hours, and a grains/milk ratio of approximately 1.0% or above. It was determined that this approach would achieve a balance between microbial activity and texture development, resulting in a lower viscosity and higher consistency. It also promoted high concentrations of lactic acid bacteria and mesophiles, leading to increased production and accumulation of organic acids in the final product. However, these higher acidity levels can alter protein interactions within the kefir's structure, potentially causing increased syneresis. This optimized combination of parameters — specifically temperature, duration and grains/ milk ratio — enables kefir manufacturers to produce a consistent, high-quality product with a profile that meets quality and probiotic standards. The resulting kefir not only meets industry standards for texture and taste, but also maintains the desired level of probiotics to ensure consumers receive the expected health benefits associated with this fermented dairy beverage.

Although the yeasts and molds in kefir were quantified, no response surface analysis could be performed on these attributes, as many results were below the quantification limit.

As a result of the optimization experiment, we identified the optimal conditions for producing high-quality kefir from pasteurized goat's milk, at a kefir grains/milk ratio of 0.9%, a fermentation temperature of 20°C and an incubation time of 24 hours. These parameters ensure the consistent production of standard kefir and improve both the quality and efficiency of the fermentation process.

3.2 Results of the changes in physicochemical, textural and microbiological attributes

Physicochemical properties

A 12-day comparative study of control kefir and kefir flavored with lemon extract revealed significant differences between the kefir types in terms of titratable acidity, proteolysis, consistency and yeasts (Table 3). In interaction with storage time, kefir type was significant for acidity, proteolysis, consistency, viscosity index, LAB and MES. Furthermore, the analysis of variance also demonstrated the significant influence of storage time on acidity, syneresis, firmness, consistency, viscosity index, LAB, MES and yeast population.

The scatter plots showed that the lemon extract influences the acidity and thus the fermentation process (Figure 6). While the ANOVA for pH showed no significant interaction ($p > 0.05$) between storage duration and kefir type, kefir type significantly impacted acidity levels. Syneresis was unaffected by type of kefir ($p > 0.05$), but was significantly impacted by storage duration ($p < 0.05$), while both kefir

type and its interaction with storage duration significantly affected proteolysis ($p < 0.05$) (Table 3).

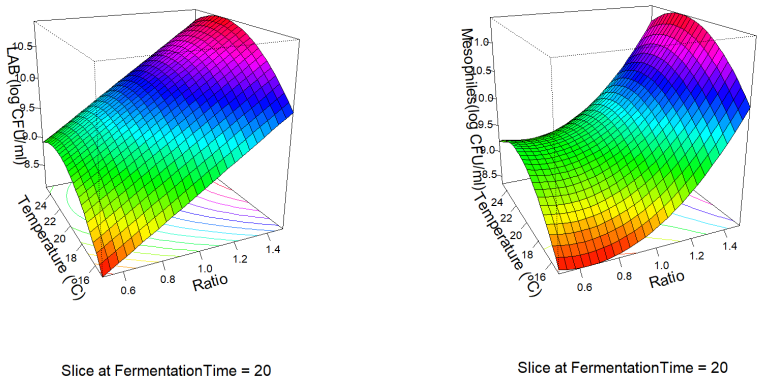

Figure 5. Response surface analysis of kefir's lactic acid bacteria and mesophiles populations (log CFU/ml), as a function of incubation temperature and grains/milk ratio, illustrating conditions that maximize microbial growth and fermentation efficiency.

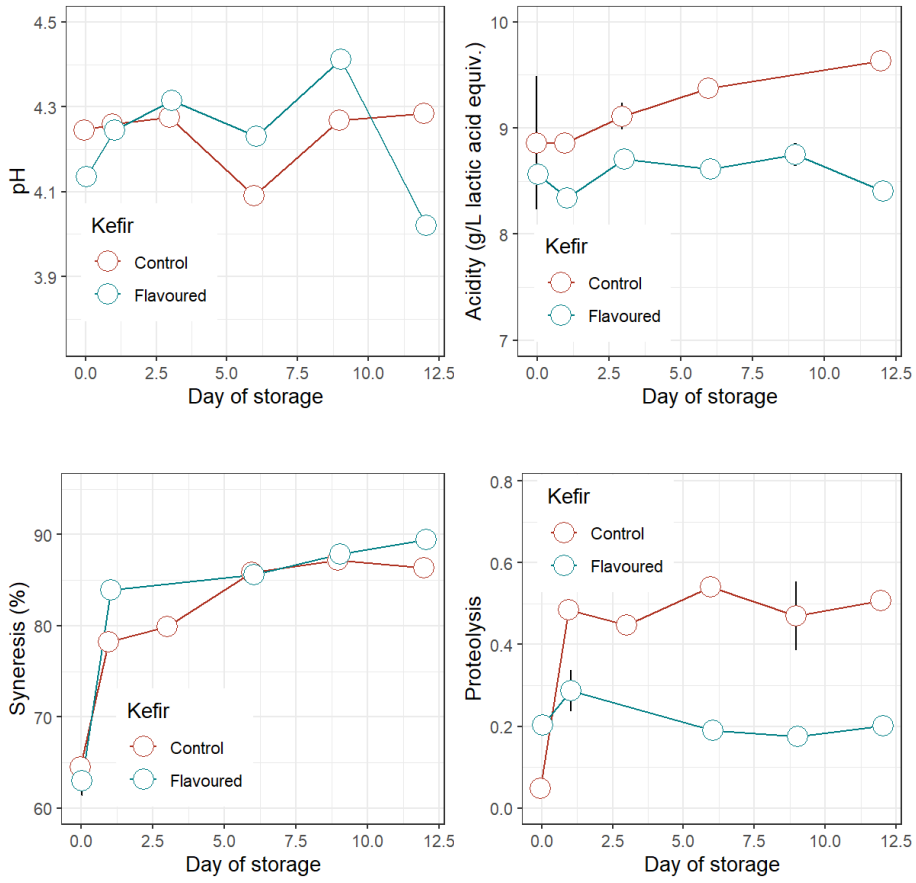

Figure 6. Changes in the physicochemical properties of goat's milk plain and flavored kefir during storage at 4 ºC

The scatter plot shows that the initial pH of the lemon-flavored kefir on day 0 was 4.12, slightly lower than the pH of the control kefir of 4.25 due to the acidity of the lemon extract. Both varieties showed a moderate increase in pH during the first three days, followed by a gradual decrease until day 6. The flavored kefir peaked at 4.4 on day 9 before dropping to 4.0 by day 12, while the control kefir remained stable at 4.28 with minor fluctuations (Figure 6). The observed pH trend in kefir aligns with the findings from Akan and Solanki et al. [15,16], who noted a decrease from 4.47 to 4.37 on day 6, followed by an increase to 4.46 on day 12. Similarly, Putri et al. reported a rise in pH of goat's milk kefir from 3.62 to 4.54 during 12 days of cold storage (6-10°C) [12]. These studies reinforced the common trend that pH increases with extended storage and highlighted its significance for maintaining kefir quality and shelf life in commercial production.

The stable pH of kefir flavored with lemon extract is likely due to the natural acidity of lemon, which initially lowers the kefir's pH. This acidity inhibits the growth of the remaining lactic acid bacteria and thus slows down fermentation. While the pH remains more stable in flavored kefir, slight decreases can still occur during storage due to residual microbial activity, though at a slower rate than in plain kefir.

In terms of acidity, the control kefir started at 8.8 g/L and increased to a peak of 9.6 g/L by day 12, compared to the flavored variant's initial acidity of 8.6 g/L (Figure 6), with the lowest acidity observed at 8.4 g/L on day 12.Wulansari et al. support the idea of microbial modulation in kefir and reported significant acidity reductions in flavored kefir over a 14-day storage period, likely due to yeast metabolism [17]. In contrast, Putri, Setiani and Warya observed a notable decline in lactic acid content in natural kefir, with acidity dropping from 2.94% to 2.46% within 16 days at slightly higher cold storage temperatures (6-10°C) [12]. These different results highlighted the complex interactions between microbial activity and storage conditions that influence kefir acidity.

Both kefir types show similar syneresis trends, starting at 63-64% on day 0 and rising to 86-89% on day 12, with the lemon-flavored kefir being slightly higher. The observed increase in syneresis during storage was consistent with Ozcan et al., who noted similar rises in samples of both plain and fruit-flavored kefir [18]. This suggests that syneresis naturally increases over time due to changes in protein structure and moisture migration.

From day 1, proteolysis of flavored kefir surged from 0.15 to 0.49 – a threefold increase. In contrast, the proteolysis of control kefir rose modestly from 0.2 to 0.29 and stabilized around 0.2. On day 12, the flavored kefir reached 0.5, underscoring the impact of the flavor additives (Figure 6). These interactions align with those of Dinkci et al., who reported comparable increases in proteolysis across various kefir samples. This highlighted a consistent trend in the change of protein degradation during storage [19].

Textural properties

A comparative analysis revealed notable differences in the texture parameters between the control kefir and lemon extract-flavored kefir over a 12-day storage period. Among these factors, only storage time had a significant impact on firmness ($p < 0.05$) (Table 3), with the control kefir starting at 11.9 g and the flavored kefir at 9.4 g, both converging to about 9 g by day 12 (Figure 7).

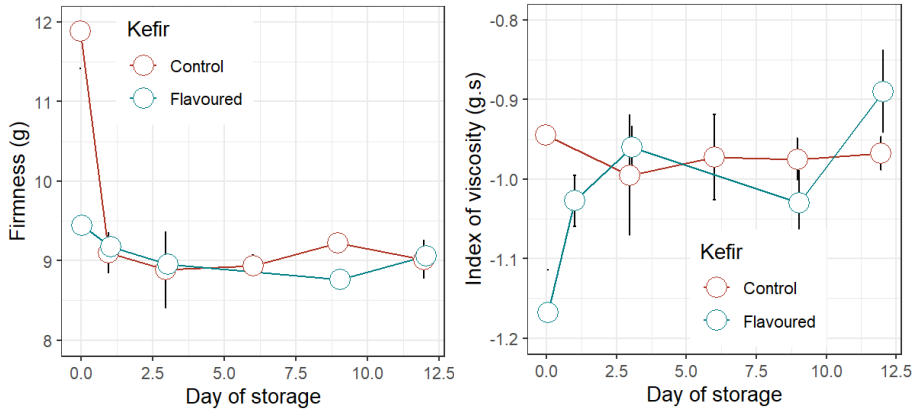

Figure 7. Changes in firmness (g) and viscosity index (g.s) of goat's milk plain and flavored kefir during storage at 4 ºC

The observed reduction in firmness is consistent with Ozcan et al., who reported a general decrease in firmness for both plain and flavored kefir samples during storage, with average values declining from 30.41 g to 19.67 g [18]. This trend highlights the impact of storage on softening the texture of fermented dairy products, attributed to ongoing fermentation and moisture redistribution, resulting in a less rigid structure over time.

The ANOVA indicated no significant effect of kefir type or storage on cohesiveness ($p > 0.05$), showing stability

despite variations in flavor. The control kefir initially had a higher cohesiveness (7.8 g) than the flavored kefir (2.9 g), but after day 1, the cohesiveness decreased in the control kefir to 4.5 g while it rose in the flavored to 5.6 g. This finding is consistent with Ozcan et al., who observed that the fruit-flavored kefir samples maintained higher cohesiveness compared to the plain kefir by the end of storage [18]. This suggests that under certain conditions, flavored kefirs can maintain or even surpass the cohesiveness of plain samples during storage.

For consistency, the control kefir started at 89 g·s compared to the flavored kefir at 70 g·s, but on day 12, the consistency declined to 55 g·s for the control kefir, while for the flavored kefir, it increased to 72.5 g·s. The results of this study differ from those of Ozcan et al., who reported decreased consistency in both plain and flavored kefir samples. Our findings suggest that lemon extract may help to maintain kefir's consistency by interacting with the microbial populations or slowing down the matrix degradation [18]. Lemon extract exhibits a broad-spectrum inhibitory effects against various microorganisms. This antimicrobial activity is attributed to several mechanisms: the disruption of bacterial membranes by the essential oils, enzyme inhibition by the flavonoids and the scavenging of free radical due to the antioxidant properties.

ANOVA also showed that storage time and kefir type significantly affected viscosity ($p < 0.05$), with the viscosity of control kefir decreasing from -0.95 g·s to -0.97 g·s, indicating structural weakening over time (Figure 7). The results of the present study on changes in kefir viscosity during storage partially align with previous research by Dinkci et al., who observed a peak in viscosity in oat milk-containing samples at the beginning of storage, followed by a decrease over time [19]. Tratnik et al. found a lower viscosity in goat's milk kefir compared to cow's milk kefir, which highlighted the influence of milk type on the viscosity [20]. Putri, Setiani, and Warya demonstrated that time-dependent viscosity increases in goat's milk kefir, emphasizing the combined effects of storage duration and temperature [12]. Their research suggested that kefir can be stored for up to 24 days without additives, with an optimal shelf life of 4-12 days, which is consistent with the timeframe of present study.

Microbiological properties

The microbial ecosystem of kefir is characterized by a variety of microorganisms, as described by Zourari and Anifantakis [21]. This complex microflora encompasses various bacterial groups, including mesophilic streptococci, Leuconostoc species and lactobacilli, which can be either mesophilic or thermophilic in nature. Additionally, the kefir microbiome is complemented by the presence of lactic acid bacteria, which are integral to kefir's fermentation process and significantly impact its flavor, texture, preservation and health benefits. These bacteria play a crucial role in product stability and contribute to kefir's unique sensory and probiotic qualities. The microbial diversity in kefir grains is extensive, with LAB species such as Lactobacillus paracasei, L. acidophilus, L. delbrueckii, L. plantarum, L. kefiranofaciens, and L. kefiri dominating the microbial population.

The statistical analysis revealed that the storage duration and its interaction with kefir type significantly affected the LAB and MES bacterial populations ($p < 0.05$) (Table 3). Initially, both the control kefir and the lemon-flavored kefir exhibited high LAB concentrations, with the control kefir at 9 log CFU/mL and the flavored kefir being slightly higher at 9.3 log CFU/mL (Figure 8). However, over a 13-day refrigerated storage period at 4°C, both types experienced a gradual decline in LAB levels, reaching 7.7 log CFU/mL in the control and 7.4 log CFU/mL in the flavored kefir by the end of the study. This reduction in LAB concentrations over time is not unexpected for refrigerated fermented products, as low temperatures slow down microbial activity. Similar results were reported by Irigoyen et al., who observed a decline in LAB populations in kefir samples between 7 and 14 days of storage [8]. However, some studies have shown contrasting results. For instance, Guzel-Seydim et al. found that the LAB in Turkish kefir remained stable and even exhibited growth after 21 days of refrigerated storage, which was possibly due to differences in microbial composition or environmental factors [22]. The study also highlighted that while lemon flavoring initially increased LAB levels, the impact of refrigerated storage at 4°C led to a general decline in LAB concentrations for both kefir types. This suggests that cold storage exerts a stronger influence on LAB stability than flavoring. These findings are significant for kefir producers who aim to preserve LAB viability in flavored kefir, as they contribute to product quality, shelf life and probiotic benefits. The research underscored the need for optimal storage conditions to maintain microbial quality and ensure the health benefits associated with kefir consumption.

At the beginning both the control and lemon-flavored kefir presented high mesophilic concentrations (9.1 and 9.3 log CFU/mL, respectively), with the flavored variant being slightly higher. However, over a 13-day refrigerated storage period at 4°C, both types experienced a gradual decline in mesophilic concentrations, with the flavored kefir showing a more significant reduction (7.2 log CFU/mL) compared to the control kefir (7.6 log CFU/mL) by day 13 (Figure 8). This faster decline in the flavored kefir might be attributed to the interaction of the lemon extract with aromatic compounds, pH modifications or other factors that create a less favorable environment for bacterial survival. These results align with research by Dinkci et al., who noted a significant reduction in Lactococcus spp. populations in refrigerated cow's milk kefir over a 21-day period [19]. The observed

decline in mesophilic populations during cold storage can be attributed to several factors, including the progressive acidification of the environment caused by the accumulation of lactic acid, competition among various microorganisms present in kefir and the gradual depletion of available nutrients. Cold storage decelerates the metabolic activity of the mesophilic bacteria without stopping it completely [23]. This allows for a slow continuation of fermentation processes even at reduced temperatures.

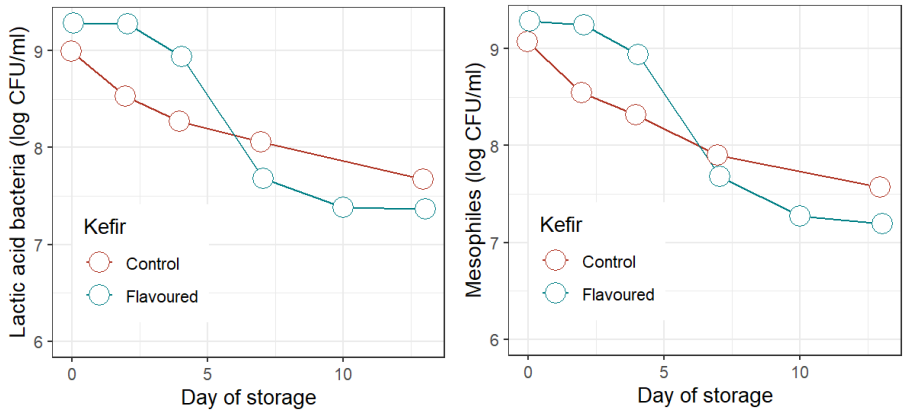

Figure 8. Changes in lactic acid bacteria and mesophiles counts (log CFU/ml) in goat's milk plain and flavored kefir during storage at 4 ºC

These findings suggest that lemon extract may act as a mild inhibitory agent on mesophilic populations, which potentially extends shelf life by reducing microbial activity while still preserving beneficial probiotic effects. Such insights are valuable for kefir manufacturers seeking a balance between flavor and microbial stability, supporting the development of products with longer shelf life and consistent quality.

The duration of samples storage did not demonstrate a statistically significant influence on the aggregate counts of yeast and mold organisms. This observation suggests that the storage duration within the tested timeframe had no substantial effect on the proliferation or reduction of these microorganisms in the tested samples. This result was in line with Setyawardani and Sumarmono, who found that the duration of storage did not significantly impact the overall yeast count in the kefir samples. After 30 days of refrigeration, the kefir maintained a yeast population of 10^5 colony-forming units per milliliter (CFU/mL). This observation suggested that the amount of yeast remained relatively stable throughout the storage period and no statistically significant changes were detected over time [24].

Yeasts, beyond lactic acid bacteria (LAB), contribute significantly to kefir's fermentation process and flavor development, especially in flavored varieties. Species from the Saccharomycetaceae family, such as Kluyveromyces and Saccharomyces, produce volatile esters and ethanol, which influence the unique aroma and flavor profile of kefir. The microbial composition undergoes dynamic changes during fermentation and storage, with different species dominating at different stages. For instance, Lactobacillus kefiranofaciens often dominates at the beginning, followed by rapid growth of Lactococcus lactis and Leuconostoc mesenteroides [25]. Storage temperature significantly affects microbial stability, with refrigerated storage (4°C) generally maintaining stable pH, acidity and microbial populations, while storage at room temperature (25°C) leads to more pronounced changes [26]. Flavored kefirs may exhibit distinct microbial profiles compared to plain varieties, as additional sugars and substrates from fruits or other flavoring ingredients (lemon extract) are added, potentially altering yeast activity and microbial interactions [27].

3.3 Sensory evaluation

Transforming goat's milk into fermented products such as kefir, especially with additional flavors or supplements, enhances both the sensory profile and the nutritional value [20]. In this study, a sensory evaluation of both plain and lemon-flavored kefir was conducted to understand consumer preferences. The panelists from the Centro de Investigação de Montanha (CIMO, Mountain Research Center) and the Instituto Politécnico de Bragança (IPB) assessed the kefirs based on appearance, odor, taste, sourness, smoothness and overall acceptability. Results of ANOVA (Table 4) revealed significant differences ($p < 0.05$) between the two kefir types in three key sensory attributes: odor ($p = 0.0041$), appearance ($p = 0.01$) and taste ($p = 0.047$). A radar or spider-web diagram (Figure 9) visually depicted the sensory profiles and showed the intensity of these attributes for each kefir type in a unified format.

Table 4. Descriptive statistics and analysis of variances of the sensory attributes of goat's milk kefir

Attribute	Control kefir		Flavored kefir		ANOVA Pr (>F)
	Mean	SD	Mean	SD	
Appearance	7.00	1.171	6.08	2.060	0.01*
Odor	5.22	1.805	6.48	2.246	0.0041**
Taste	4.23	2.034	5.10	2.059	0.047*
Sourness	5.09	1.87	5.45	1.698	0.34
Smoothness	5.40	2.012	5.56	2.050	0.71
Acceptance	4.76	2.17	5.58	1.972	0.062 .

'***': p-value between 0 and 0.001 (highly significant)
'**': p-value between 0.001 and 0.01 (very significant)
'*': p-value between 0.01 and 0.05 (significant)
'.': p-value between 0.05 and 0.1 (marginally significant)
' ': p-value greater than 0.1 (not significant)

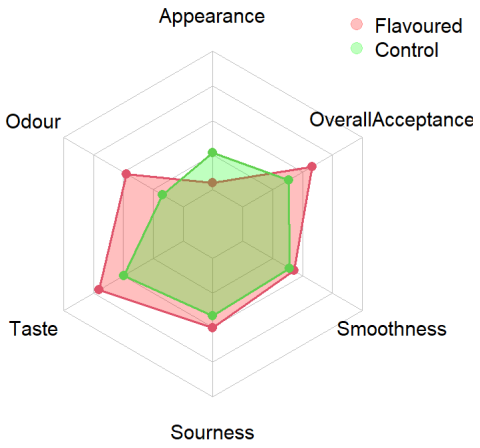

Figure 9. Spider web plot of the respondents' appreciation of kefir

Panelists rated the odor of the flavored kefir significantly higher than that of the control, suggesting that the lemon extract may effectively mask or soften the strong "goaty" aroma typical of goat's milk kefir, making it more appealing to the consumers. This finding underscored the potential of flavoring as a valuable tool to improve the sensory appeal of goat's milk kefir. Furthermore, the flavored kefir scored higher in taste, which can be attributed to the refreshing citrus notes from lemon extract that balance and complement the sourness of kefir, thereby improving its overall palatability. These results collectively support the use of natural flavor additives to expand kefir's flavor profile and increase its appeal. With regards to appearance, the plain kefir received higher scores than the flavored type ($p = 0.01$), likely due to its unaltered, natural kefir appearance, which some consumers associate with authenticity and freshness (Figure 9).

Interestingly, the overall acceptability ratings of the kefir samples approached the significance ($p = 0.062$), indicating a mild but notable preference for the flavored kefir. While the ratings for sourness and smoothness were statistically similar between the two kefirs, the enhanced odor and taste made the flavored variant more appealing to panelists (Table 4). Irigoyen et al. observed a high acceptability of kefir samples during the initial days of storage [8]. Additionally, the panelists favored attributes such as milky taste, pleasant odor, and balanced viscosity, which resonate with earlier research by Muir, Tamime, and Wszolek, who identified these factors as crucial for consumer satisfaction with fermented dairy products such as kefir [28]. Thus, the sensory evaluation shows that lemon-flavored kefir is well accepted, as indicated by the overall acceptability assessment. These findings highlight the potential of flavors to improve the appeal of goat's milk kefir, make it more acceptable to consumers and expand its market potential.

As for sourness, the flavored kefir was noted for its stronger acidic fragrance — a characteristic often favored in fermented dairy products. Supporting these findings, Arroum et al. emphasized that acidity, flavor, taste and odor are sensitive to dosage and fermentation time, each contributing meaningfully to consumer perceptions of kefir's acceptability [29]. Flavoring can be particularly

beneficial for goat's milk kefir. According to Tratnik et al., fermenting goat's milk with kefir grains can help to mask the characteristic "goaty" flavor, which some consumers find unpleasant, and thus make the product more palatable [20]. The findings of this study align with these observations, as the lemon-flavored kefir was perceived as more balanced in taste and odor due to the masking effect of lemon extract. Moreover, kefir grains significantly influence the sensory quality by shaping the texture and consistency, which affect the creaminess and smoothness [30]. In summary, the sensory evaluation demonstrated that lemon-flavored kefir holds a distinct advantage in consumer acceptance, receiving high ratings for key attributes such as odor, taste and overall acceptability. These results highlight the potential of natural flavoring agents such as lemon extract to enhance sensory attributes and increase the appeal of kefir to consumers. This insight is valuable for product development as it suggests that strategic flavor modifications could elevate the marketability of goat's milk kefir and make it more attractive to a wide range of consumers.

4. Conclusion

This study aimed to explore the relationship between kefir's shelf life and its physicochemical and microbiological composition under different conditions and to identify the key factors that affect quality over time. In the first phase of the study, we established optimal production parameters and determined a kefir grains/milk ratio of 0.9%, a temperature of 20°C and an incubation time of 24 hours as ideal for producing standard kefir from pasteurized goat's milk. With this combination, desirable physicochemical criteria, such as low syneresis and high acidity, along with appropriate textural and microbiological standards were achieved. The second phase revealed significant differences between the flavored and control kefir in their quality properties during shelf life; flavored kefir maintained a lower pH due to the activity of the lactic acid bacteria, which contributes to the inhibition of pathogens. While both kefir types exhibited similar firmness, the flavored kefir had a higher viscosity index and consistency. Our findings have practical applications for industrial-scale kefir production, as the optimal fermentation temperature balances product quality and efficiency and potentially reduce production costs. Understanding how acidity evolves during storage can help producers adjust initial levels to maintain a consistent flavor, while the relationship between temperature and viscosity allows for texture customization to meet consumer preferences. By applying these optimized parameters, kefir producers can improve process efficiency, enhance product consistency, and boost consumer appeal, making these findings valuable for commercial production. The sensory analysis revealed a distinct preference for flavored kefir, which was rated superior in odor, taste, and overall acceptability than the plain kefir. This underscores the positive impact of the lemon extract on the sensory attributes of kefir. Overall, this study provided valuable insights into the composition and storage stability of natural versus flavored goat's milk kefir. It highlights avenues for future research on its nutritional benefits and optimal packaging solutions to enhance products' longevity. Future studies could explore the health benefits of flavored versus plain kefir through clinical trials, investigate alternative flavors such as berries or herbs to assess their impact on microbial diversity and consumer preferences, and examine kefir's long-term stability under varied storage conditions. These studies could drive product innovation, improve shelf life and enhance consumer appeal.

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Authors' contribution

M.A.: Validation; Investigation; Visualization; Data curation; Writing—original draft preparation; Writing—review and editing; S.Y: Writing—review and editing; Validation; Visualization; Supervision; V.C: Conceptualization; Methodology; Software; Validation; Resources; Writing—review and editing; Supervision; Project administration; Funding acquisition; U.G-.B: Conceptualization; Methodology; Software; Validation; Resources; Data curation; Writing—original draft preparation; Writing—review and editing; Supervision; Project administration; Funding acquisition.

Availability of data and materials

Data sharing is not applicable to this article.

Declarations

Not applicable.

Consent for publication

All authors have read and agreed to the published version of this manuscript.

Conflicts of interest

Not applicable.

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