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**Development of kefir from goat milk with guava pulp**

**Desenvolvimento de kefir a partir de leite de cabra adicionado de polpa de goiaba**

**Desarrollo de kéfir a partir de leche de cabra añadida con pulpa de guayaba**

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**Abstract**

Goat milk may be an alternative to cow milk to people that suffer from allergic conditions, besides presenting high digestibility. Among dairy products, fermented milk is one of the most consumed. In this category, Kefir is a fermented milk that presents functional properties. Therefore, this laboratory research aimed to obtain Kefir from goat milk added by different guava pulp concentrations (0, 5, 15 and 25 %) and evaluate its physicochemical, microbiological and sensorial characteristics through quantitative data. Data obtained from microbiological analysis evidenced food safety for consumers and probiotic potential. The pulp concentration variation changed some physicochemical parameters. Sensorially, all the samples had high positive answer levels in the acceptance test, besides the last group had high percentage of buying intention. This study demonstrates that Kefir added with the higher guava pulp concentration presents potential to be introduced in the Brazilian market.

**Keywords:** Fermented milk guava; Dairy product; Characterization; Tropical fruit.

**Resumo**

O leite de cabra pode ser uma alternativa ao leite de vaca para pessoas que sofrem de condições alérgicas, além de apresentar alta digestibilidade. Entre os laticínios, o leite fermentado é um dos mais consumidos. Nesta categoria, o Kefir é um leite fermentado que apresenta propriedades funcionais. Portanto, este trabalho laboratorial objetivou produzir Kefir a partir de leite de cabra com diferentes concentrações de polpa de goiaba (0, 5, 15 e 25%) e avaliar suas características físico-químicas, microbiológicas e sensoriais através de dados quantitativos. Os dados obtidos da análise microbiológica evidenciaram a segurança alimentar dos consumidores e o potencial probiótico. A variação da concentração de polpa alterou alguns parâmetros físico-químicos. Sensorialmente, todas as amostras apresentaram altos níveis de resposta positiva no teste de aceitação, além do último grupo ter alto percentual de intenção de compra. Este estudo demonstra que o Kefir adicionado com a maior concentração de polpa de goiaba apresenta potencial para ser introduzido no mercado brasileiro.

**Palavras-chave:** Leite fermentado; Goiaba; Produto lácteo; Caracterização; Fruta tropical.

**Resumen**

La leche de cabra puede ser una alternativa a la leche de vaca para las personas que padecen afecciones alérgicas, además de tener una alta digestibilidad. Entre los productos lácteos, la leche fermentada es una de las más consumidas. En esta categoría, el kéfir es una leche fermentada que tiene propiedades funcionales. Así que esta investigación de laboratorio tenía como objetivo producir kéfir a partir de leche de cabra con diferentes concentraciones de pulpa de guayaba (0, 5, 15 y 25%) y evaluar sus características físico-químicas, microbiológicas y sensoriales a través de datos cuantitativos. Los datos obtenidos del análisis microbiológico mostraron la seguridad alimentaria de los consumidores y el potencial probiótico. La variación en la concentración de pulpa cambió algunos parámetros físico-químicos. Sensorialmente, todas las muestras mostraron altos niveles de respuesta positiva en la prueba de aceptación, además del último grupo que tenía un alto porcentaje de intención de compra. Este estudio demuestra que el kéfir agregado con la mayor concentración de pulpa de guayaba tiene el potencial de ser introducido en el mercado brasileño.

**Palabras clave:** Leche fermentada; Huayaba; Producto lácteo; Caracterización; Fruta tropical.

**1. Introduction**

Kefir is a fermented milk from native microbiota of polysaccharide and proteins structured grains. Among the diverse microorganisms that compose that, yeasts, acetic and acid lactic bacteria are found, which promote benefits to the human intestinal regulation, as well as immunological and antagonistic effects, causing an increase in resistance against pathogens (Costa et al., 2013, Santos & Basso, 2013). These microorganisms also provide biological safety, due to its acid lactic and other compounds production, acting as a natural preservative that gives higher stability to the product (Weschenfelder et al., 2011).

Allying the trend for healthy products to desired functional properties and sensorial characteristics, the food industry has been directing efforts to meet the consumer market demand. In this context, goat milk may be used in Kefir production, which may be consumed “in natura” or added with fruits.

The goat milk is a very consumed product in a lot of countries around the globe, but in Brazil, its’ consuming is still low due to the consumers’ resistance to its characteristic smell, distinct from cattle milk, which is due to the presence of caprylic acid and short chain fatty acids (Borges, 2006).

However, goat milk has a lot of benefits, such as smaller fat globules, which ease the product’s digestion; smaller contents of α-S1 caseín, which makes it hypoallergenic, besides presenting nutritional value similar to cattle milk. This way, the goat sourced product attends the nutritional needs of people who suffer from cow milk allergy or other gastrointestinal disorders (Haenlein, 2004).

Fruit addition makes goat milk Kefir more attractive sensorially and richer in nutrients (Almeida et al., 2011), guava, for example, has good public acceptance for “in natura” consumption, besides high nutritive properties (Scremim, 2007).

Given the above, this work had for goal to make and evaluate the physical-chemical, microbiological and sensorial features of goat milk Kefir added by different concentrations of guava pulp.

**2. Methodology**

The present work can be defined as a quantitative laboratory research, whose methodology measures qualities in numerals in its due units, generating data that can be analyzed mathematically and statistically (Pereira, 2018).

The Kefir was produced in the Dairy Laboratory of the Agrarian Science Center – (CCA) of the Federal University of Ceará, using goat whole UHT milk, whom was fermented with Kefir grains previously activated in B.O.D. at 25 ºC for 24 hours. In this period milk homogenization was performed five times at two hours intervals. After the fermentation, the product maturated in a fridge at 10 ºC for 24 hours. Then the Kefir grains were taken from the milk with a sieve. The obtained product was added by saccharose (10% m/v) and guava pulp in different concentrations: 0% (A), 5% (B), 15% (C) and 25% (D). Homogenization occurred manually, for one minute, with a glass stick. The whole process can be seen in Image 1.

**Image 1.** Goat milk Kefir added by guava pulp elaboration fluxogram.



 Source: Author (2020)

On the image above it’s possible to observe the kefir production and its inputs. For analysis, the goat milk had the pH (AOAC, 2001), Dornic acidity, density at 15 ºC, fat content and cryoscopy index evaluated, according to methodology indicated by Institute Adolf Lutz (IAL, 2008).

 The pasteurized guava pulp was submitted to total solids analysis using an analogical refractometer model N-1α, Atago.

 The physical-chemical analysis of the goat milk Kefir beverage added by guava pulp were performed at the laboratories of the Department of Food Engineering, Agrarian Science Center - CCA, Federal University of Ceará - UFC, consisting in: protein by the Kjeldahl method (AOAC, 2001), total solids, ashes and fat content, pH and titrable acidity following methodology suggested by Institute Adolf Lutz (IAL, 2008). Viscosity was measured at the Chemistry Laboratory of the Federal Institute of Ceará – IFCE through viscometer, model DV2T, at 10 ºC. The color of the products was analyzed though the CIELAB system, using Konica Minolta CR410 colorimeter.

 The sensorial acceptance test was performed through 9-points hedonic scale (9: like extremely, 5: neither like nor dislike, 1: dislike extremely) to evaluate the features of appearance, smell, flavor, consistency, color and global impression. For the acidity, consistency and guava flavor ideality test, a 9-point scale was also used: (+4: extremely stronger than the ideal, 0: ideal, -4: extremely weaker than the ideal). Purchase intention was evaluated through a 5-point scale (5: certainly would buy, 3: not sure if would buy; 1: certainly wouldn’t buy). Those analyses were performed by 72 not trained testers, both feminine and masculine genders, between 18 and 65 years old. Such analyses took place at the Sensorial Evaluation Laboratory at CCA, UFC.

 The results of the physical-chemical and sensorial analysis were treated statistically through the Variance Analysis ANOVA in order to find the statistical significance of the results. The averages obtained for the samples were compared through Tukey test, with significance level at 5%. The software used for data processing was ASSISTAT 7.7.

**3. Results and Discussion**

The obtained results from the goat milk (Table 1) are according to the established standards of the current legislation from the Brazilian Ministry of Agriculture, as stated by the Technical Regulation of Production, Identity and Quality of Goat Milk (Brasil, 2000).

**Table 1.** Average results (± standard deviation) of the physical-chemical properties of the goat milk.

|  |  |
| --- | --- |
| **Parameter** | **Result** |
| Titrable acidity (g lactic acid/100 g sample) | 0,18 ± 0,00 |
| Density (g/ml, 15 ºC) | 1,032 ± 0,002 |
| Cryoscopy index (ºH) | -0,550 ± 0,003 |
| Fat (%) | 3,20 ± 0,07 |

Source: Author (2019).

Therefore, note that the product developed was produced within Brazilian current law statements with the purpose of health and safety for consumers.

The value for the soluble solids found at the pasteurized guava pulp (Table 2) was according to the General Technical Regulation for Identity and Quality Standards for Fruit’s Pulp (BRASIL, 2000).

**Table 2.** Total soluble solids analysis of the pasteurized guava pulp.

|  |  |
| --- | --- |
| **Parameter** | **Guava Pulp** |
| Total soluble solids (ºBrix) | 7,07 ± 0,06 |

Source: Author (2019).

Nevertheless it is safe to conclude that the guava pulp was safe for use and according to Brazilian current laws for consumers’ health and safety.

Sample A (Control) presented higher pH value than the other samples, presenting significant difference (p<0,05), however, its acidity was the highest. Such situation may be explained by the greater percentage of proteins in sample A. This macronutrient may have acted as a buffer, preventing pH changes. Temiz and Kezer (2015) performed substitution of fat for a protein-based substance in a Kefir obtained from a cattle’s and goat milk mix, which would cause a buffer effect on the samples.

 According to Almeida et al. (2011), goat milk Kefir shows acidity higher than the cattle milk Kefir. In this parameter, samples A and D differed significantly (p<0,05) when compared between each other and among the rest. The lactic acid quantity was superior at sample A (Control), suggesting that the guava pulp interfered in acidity. Data obtained by Nurliyaniet et al. (2015) indicate acidity value for goat milk Kefir as 0,93±0,15, inferior to that determined for the control sample, probably, due to the fermentation period being shorter than the one performed in the present work (24h).

 According to the Technical Regulation of Identity and Quality for Fermented Milks (BRASIL, 2007) all samples had acidity values within the proper standard (0,5 – 1,5).

 Sample D, added with the higher pulp concentration (25%), presented less fat and protein content when compared to sample A. The protein (1,1%) and fat (insignificant) values from the pasteurized guava pulp (Petruz fruity, 2017) did not interfere with the protein and fat content of Kefir. The percentage of protein found at the control sample was similar to the fermented milk added by probiotics evaluated by Cunha (2008).

 The average comparison for the ashes parameter did not point to any significant difference (p<0,05) among the samples. Such results are similar to the ones found at the Kefir samples added by an alcoholic vegetal extract in the work of Zelovitls et al. (2016).

 The result from the guava pulp total solids analysis is inferior to that from the control (Sample A), thus, the samples with higher pulp concentration (C and D) showed a lower solids value than the other samples.

 Samples C and D presented significant difference for viscosity when averages were compared among each other. Therefore, viscosity increased according to the guava pulp concentration increase at Kefir (Table 3).

**Table 3**. Average values (± Stardard deviation) for the Kefir from goat milk added by guava pulp characterization. A: 0% Pulp; B: 5% Pulp; C: 15% Pulp; D: 25% Pulp.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Parameter** | **A** | **B** | **C** | **D** |
| **pH** | 3,93 ± 0,01a | 3,89 ± 0,01b | 3,89 ± 0,01b | 3,90 ± 0,01b |
| **Titrable acidity (% Lactic acid)** | 1,19 ± 0,05a | 1,10 ± 0,01b | 1,04 ± 0,03b | 0,91 ± 0,02c |
| **Total soluble solids (ºBrix)** | 17,0 ± 0,8a | 17,3 ± 0,4a | 14,8 ± 0,4b  | 15,6 ± 0,3b |
| **Ashes (%)** | 0,66 ± 0,01a | 0,71 ± 0,02a | 0,69 ± 0,02a | 0,68 ± 0,02a |
| **Fat (%)** | 3,2 ± 0,1a | 3,1 ± 0,5ab | 3,0 ± 0,5ab | 2,6 ± 0,01b |
| **Protein (%)** | 2,6 ± 0,10a | 2,4 ± 0,11ab | 2,26 ± 0,05b | 2,05 ± 0,01c |
| **Viscosity (cP)** | 5,55 ± 0,18c | 10,02 ± 0,25c | 23,43 ± 0,18b | 54,38 ± 0,11a |
| **L\*** | 84,97 ± 0,09a | 83,99 ± 0,07b | 81,34 ± 0,04c | 75,8 ± 0,05d |
| **a\*** | - 3,08 ± 0,03a | - 0,87 ± 0,07b | 2,94 ± 0,05c | 7,28 ± 0,02d |
| **b\*** | 8,61 ± 0,08a | 7,52 ± 0,05b | 7,30 ± 0,02c | 7,41 ± 0,02bc |

Source: Author (2019)

Scremin (2007) verified that the viscosity of pasteurized ripe guava pulp is high, therefore, the samples with higher pulp concentration showed higher viscosity than the control sample. Note that samples C and D presented positive color results for parameter a\*, indicating tendency to red. All samples showed positive values for parameter b\*, suggesting yellow bias. Sample A had the most elevated value for the parameter of luminosity (L\*), possibly, due to its higher goat milk quantity, which contains Vitamin A in its active form, giving to the product a whitish coloring, according to Pellegrini (2012).

The values obtained through the microbiological counting (Table 4) obeyed the current legislation from Ministry of Agriculture, as seen in the Technical Regulation of Identity and Quality of Fermented Milks (Brasil, 2007), which proves the beverage’s food safety.

**Table 4.** Results from the microbiological analysis of the goat milk Kefir added by guava pulp.

|  |  |  |  |
| --- | --- | --- | --- |
| **Samples** | **Coliforms at 45 ºC (MLP/g)** | **Total acid lactic bacteria (FCU/g)** | **Molds and yeasts (FCU/g)** |
| A | < 3,0 | 2,2 x109 | 1,0 x105 |
| B | < 3,0 | 5,0 x 109 | 3,5 x 106 |
| C | < 3,0 | 4,5 x 109 | 7,5 x 106 |
| D | < 3,0 | 3,6 x 109 | 7,0 x 106 |

Source: Author (2019).

It can be seen that all the samples showed above have lactic bacteria counting above 109, pointing to a probiotic potential. The obtained values for molds and yeasts and for coliforms at 45 ºC were similar to those found by Santa et al. (2008).

Sensorial evaluation rated the samples’ acceptability through a 9-point hedonic scale for sensorial properties and a 5-point scale for consumers’ buying intention. Data show that all samples were at the acceptance zone (> 50%; between 6 and 9 at the scale) for parameters of color, appearance, flavor, smell, consistency and global impression, as seen in Table 5.

**Table 5.** Results from the sensorial evaluation of the goat milk Kefir added by Guava pulp, displaying the average through hedonic scale.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Parameter** | **Sample A** | **Sample B** | **Sample C** | **Sample D** |
| Color | 6,01 ± 1,939a | 6,56 ± 1,767a | 6,24 ± 1,888a | 6,40 ± 1,692a |
| Appearance | 5,63 ± 1,850a | 6,11 ± 1,850a | 5,60 ± 1,881ab | 5,76 ± 1,917b |
| Smell | 6,38 ± 1,989a | 6,99 ± 1,873b | 5,74 ± 1,972bc | 6,78 ± 1,786c |
| Flavor | 5,83 ± 2,332a | 6,03 ± 2,188a | 5,44 ± 2,257a | 5,74 ± 2,245a |
| Consistency | 5,85 ± 1,859a | 5,82 ± 1,771a | 6,82 ± 8,476a | 5,68 ± 1,865a |
| Global impression | 5,86 ± 1,802a | 6,35 ± 1,721ab | 5,54 ± 1,838bc | 6,03 ± 1,776c |
| Buying intention | 2,78 ± 1,091a | 3,31 ± 1,158ab | 2,54 ± 1,278bc | 2,94 ± 1,112c |

Source: Author (2019).

It is possible to observe in the table above that sample D exhibited higher percentage of grades at acceptance zone for color, appearance and global impression. Samples B and D presented higher testers acceptance for flavor and consistency. In Schimidt et al. (2012), 69 % of the interviewed testers said that the flavor is the most utilized criterion during dairy products buying, results that match with the observed, once sample D, the best rated sample for guava flavor, has the higher buying intention index.

The ideality test was performed through a 9-point scale, whose grades below zero would point to a less than ideal feature, equals zero would be ideal, and grades higher than zero would show an above ideal feature. In this test, as seen in Image 2, Sample D also excelled at it, reaching high percentage of ideality for acidity and guava flavor.

**Image 2.** Results from ideality test performed with goat milk Kefir added by guava pulp, where negative grades indicate features below ideal, positive grades indicate above ideal features and zero means ideality. A: Control sample; B: 5% Guava Pulp; C: 15%; Guava Pulp; D: 25% Guava Pulp,

Source: Author (2020)

Therefore it is possible to conclude that sample D is very well received by the target audience, mainly in comparison to control sample and samples with less guava pulp.

**4. Final Considerations**

 Through the present work it is possible to conclude that the goat milk kefir added by guava pulp was developed successfully, being well received by the public and, when characterized, showed properties within current Brazilian legislation. Products like these add variety in the market and take innovation to consumers’ tables.

Physical-chemical features allowed us to observe that the pulp concentration influences acidity, protein content, viscosity and color of the goat milk kefir.

Microbiological results point that all samples have probiotic potential, bringing benefits to the consumer. However, sample D outstands for its high consumers’ acceptance, besides having a positive buying intention and thus a high market potential.

 For following works, shelf-life shall be evaluated along with different possible packages and store settings, so that such a product may be commercialized with all its variables characterized.

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