




Development of milk drink with whey fermented and acceptability by children and adolescents

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Revised: 21 January 2021 / Accepted: 26 January 2021
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Abstract Malnutrition is one of the main trouble relationship children development, it is linking of infectious disease, affect brain development, learning delay and others. The school feeding is an important action to mitigate this, but the acceptability of new and healthy products still a challenge. The goal of this article was to develop a fermented drink, replacing milk to whey, added of mangaba pulp and iron, to improve the nutritional quality of products. Two formulations were developed with a difference in ratio milk and whey. The pH, acidity, nutritional label, microbiology and sensorial assay with target public (children and adolescents) of formulations were evaluated. Highlights the contribution of protein, calcium and iron to daily value of intake recommended of formulations, 8.4%, 15.2% and 44.3%, respectively. The microbiological parameters founded shows that the formulations were developed according to good manufacture practice. The formulation and age showed significant effect in acceptability of judges, but gender did not effect. The increase of whey concentration in formulation improved the acceptability, ranking to 91.5% to children and 73.6% to adolescents. The developed formulations are a great option to novel food products, given the high acceptance of the

fermented milk drink by potential consumers and nutritional aport.

Keywords Malnutrition · School feeding · Whey · Acceptability

Introduction

Malnutrition refers to deficiencies or excesses in a person's intake of energy and/or nutrients, and it is widely distributed among people and ages, but occurs mainly in children. The World Health Organization reveals that 49 million children under 5 years of age are wasted while 40 million are overweight or obese (Kulkarni 2016). These results become the malnutrition a world health crisis and one of United Nations' Sustainable Development Goals (SDGs).

The synergistic relation between malnutrition and other disease is well known, mainly infectious disease (Rice et al. 2000; Bhutta et al. 2017). In addition, a wide range of cognitive deficits were observed in malnourished children, and it is associated with motor and language development. The consequence of this is a delay on intellectual development and learning disabilities in school children (Jomaa et al. 2011).

To mitigate this problem, school feeding programs aim to enhance the concentration and learning capacity of school children by providing a meal in school to reduce hunger that may otherwise impair children's performance (FAO 2005). Milk and dairy products are one of the products can be offered in school feeding, and can play a particularly important role in human nutrition, due the energetic supply and minerals and vitamins sources (Stobaugh et al. 2016). The use of whey in food development is

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an option for increase a nutritional and is an effective cheaper alternative to the standard milk (Bahwere et al. 2014; de Souza et al. 2020). The functional properties of fermented milk can be increased by including fruits, nuts, and herbs to meet the increasing consumer demand (De Andrade et al. 2019). Brazil is one of the three major fruit producers in the world and native fruits play an important role in the economy locally (Clerici and Carvalho-Silva 2011). Mangaba (*Hancornia speciosa* Gomes) stands out for the nutritional profile, such as total phenolics, vitamins, carotenoids, and minerals (Schiassi et al. 2018). The fruit processing into products contribute to economical sources of functional and healthy ingredients of diets.

Even if a product has been developing to offer the suitable nutritional aport, the acceptability of consumers is crucial, mainly children and adolescents. According to Paiva et al. (2016), the acceptability of school meals that are offered is still a challenge. The good acceptability of health-food can contribute to the development of healthy food lifestyles (Lülfs-Baden and Spiller 2009; Oostindjer et al. 2017). The goal of this study was to develop a fermented milk drink using whey, to indicate to feeding school a high-quality alternative beverage for increase the nutritional value and that possesses good acceptance among children and adolescents.

Material and methods

Materials

Whey and milk UHT was purchased from the local market. For the fermentation of milk drink, a commercial lyophilized starter culture containing mixed culture of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* ssp *bulgaricus* were used (YoFlex Harmony 1.0–C.H, Hørsholm, Denmark). The mangaba (*Hancornia speciosa* Gom.) fruits were obtained from Cooperativa Grande Serão (Montes Claros, MG, Brazil). Ingredients used in formulation were purchased from local market. All reagents used in this study were analytical grade.

Methods

Preparation of mangaba's pulp

Initially, the fruits were visually selected. Undesirable fruits were discarded while intact, ripened, firm ones were washed and dipped in water containing 30 ppm of sodium hypochlorite for 15 min. Then, mangaba was depulped, filtered through a 0.25-mm-mesh stainless steel sieve in a de pulper (Bonima®) and pasteurized at 65 °C for 30 min.

The pulp was frozen at -20 ± 2 °C until being thawed for formulation.

Incubation of starter culture

The starter culture was previously incubated separately in 1% (v/v) of sterilized skimmed milk powder diluted to 10% (w/v). A 2:1 ratio was used in inoculation, adding up to 1% of the total volume of the portions. After inoculation, the material was incubated at 45 °C in bacteriological culture chamber until coagulation.

Development of milk fermented drink

Two formulations were developed, differed in the milk-to-whey ratio: F1 contained 70% (v/v) of milk and 30% (v/v) of reconstituted whey (in water—15%), whereas F2 had 50% (v/v) of milk and 50% (v/v) of reconstituted whey (in water—15%). The other ingredients were same in both formulations: sugar (12% w/v), mangaba pulp (5% w/v), lactic acid culture (1% v/v), modified starch (0.8% w/v) and iron amino acid chelate (3 mg/100 mL). Iron addition to both formulations was standardized according to Brazilian legislation (BRASIL 2005a).

In this process, the mixture of pasteurized milk, reconstituted whey, sucrose and modified starch was subject to heating at 75 °C for 15 s. Then the starter culture was added at 45 °C. The fermentation was finished when pH reach of 4.6 (6–8 h) and then cooled to 4 ± 2 °C, temperature which was kept throughout clot breaking, homogenization after supplementation with mangaba pulp and iron. The fermented milk drink were packed in one-liter plastic bottles and stored in a refrigerator at 5 ± 2 °C until analyses.

pH and titrable acidity

The pH of milk fermented samples was measured at room temperature (25 ± 2 °C) by potentiometry (INSTRUTHERM, Brazil). The titrable acidity was determined after homogenization of sample, and 10 mL of sample was diluted in 10 mL of distilled water and titrated with NaOH solution (0.1 M). The results were expressed in g of lactic acid/100 mL of sample (AOAC 2000).

Development of nutritional label

The nutrition label of formulations developed was elaborated according to Brazilian Food Composition Table (TACO 2006). The % daily value (DV) was calculated based on Brazilian legislation (BRASIL 2005b) and dietary reference intake (FAO/WHO Expert Consultation 2005).

Microbiological analysis

Microbiological analysis of formulations (F1 and F2) was performed by complete homogenization of 25 mL of sample with 225 mL of peptone water (0.1%), followed by serial dilution and seed in specific media for the determination of different microorganism groups.

Total coliform determination presumptive test was conducted by inoculating into 3-tube series containing Lauryl Sulfate Broth (Oxoid) and incubated at 35 ± 2 °C for 48 ± 3 h. Samples with positive presumptive outcomes were transferred to Brilliant Green Bile (Oxoid) broth and incubated at 35 ± 2 °C for 24 to 48 h. Results were expressed as most probable number (MPN) g^{-1} . For yeast and mold counts were streaked on potato dextrose agar medium (PDA) acidified (pH 3.5) and incubated at 25 °C for 5 d. The results were expressed as colony-forming units (CFU) g^{-1} (APHA 2015).

Sensory evaluation

Before starting the sensory evaluations, the experimental protocol was analyzed and approved by the institutional Research Ethics Committee (Process no 0465.0.203.000-10) of Federal University of Minas Gerais. Students of high school were judges of this study. Previously, their parents or caregivers approved their participation through Informed Consent Term. Students untrained and possible potential consumers of the product evaluated the samples using a 5-point facial hedonic scale. The samples offered separately to judges. A numerical value was assigned for each illustration on the hedonic scale: the picture corresponding to a facial expression of “dislike extremely” was assigned the numerical value of 1, while the picture corresponding to the facial expression “like extremely” was assigned the value 5. The index of acceptability (IA) was calculated using Eq. 1:

$$IA(\%) = \frac{Score}{5} \times 100$$

Statistical analysis

Analysis of variance (ANOVA) and randomized block design were used. Data were analyzed in the System Statistical Analysis (SAS) software, version 9.0, by F test at 5% of significance level. To identify the formulation with the highest acceptability, Tukey test was applied at 5% of significance level.

Results and discussion

pH and acidity

The evaluation of pH and acidity of a fermentative process is essential for the maintenance of viable cells. The final pH of F1 was 4.17 and 4.2 to F2, both with an adequate pH range (3.6 to 4.6) as to post-processing lactic bacteria maintenance. The acidity value of both formulation were 0.61%, this results showed low acidifying characteristic of starter lactic culture in milk fermentation, since the acidity for fermented milk products ranges from 0.7 to 0.9% of lactic acid (FDA 1982). Souza et al. (2020) evaluated the stability of dairy milk drink with caja-mango fruit fermented with same lactic culture, among 14 days, and observed ranches pH 4.18–4.31 and acidity of 0.81–0.80%. According the authors, the maintenance of lactic acid level can be due reduction of lactose during fermentation, causing the non-variation of acidity of product.

Although information on minimum effective concentrations of viable cells were not established during or the end of fermentation, it is generally accepted that probiotic products should have a minimum concentration of 10^6 (CFU) g^{-1} (Kechagia et al. 2013). The results shows that the conditions of fermentation are favorable to maintenance of viable cells, suggesting that this products developed is a potential probiotic drink.

Nutritional analysis

Table 1 shows the proximate compositions of F1, F2, and mangaba pulp, as well as their respective equivalences to daily nutritional values and RDI of the components present in the formulations. F1 and F2 showed approximately the same carbohydrate content because they presented similar concentrations of this macronutrient originated from lactose in milk and whey, regardless to the milk-to-whey ratio. Also, we added the ingredients that deliver carbohydrates to the formulations (mangaba pulp, modified starch, and sugar) in identical concentrations.

Both formulations presented fibers, which are attributed to the supplementation with mangaba pulp (Clerici and Carvalho-Silva 2011). Fibers are vegetal food components with high functional activity that are capable of remarkably increasing satiation, retarding digestion, absorbing fatty acids and glucose, and contributing to stool formation and to lessening stool residence time in colon (Jones 2014).

All formulation showed daily values to minerals (Ca and Fe) upper to 14%, indicating that these minerals can be declared on the label. The % of daily value intake of calcium decreased in F2 formulation, 15%. This is explained by the different milk-to-whey ratio used in both

Table 1 Nutrition information of the fermented milk developed and mangaba's fruit pulp contribution

Nutrient	F 1	F 2	M	RDI	% DV 1	% DV 2	% DV M
<i>Nutrition information (200 mL)</i>							
Energy value (kcal)	196.8	183.4	132.4	2000.0	9.8	9.2	6.6
Carbohydrate (g)	33.8	33.5	20.0	–	–	–	–
Protein(g)	4.6	3.8	2.4	50.0	9.2	7.6	4.8
Total fat (g)	4.8	3.8	4.8	78.0	6.2	4.9	6.2
Total Fiber (g)	0.4	0.4	6.8	25.0	1.6	1.6	27.2
Ca (mg)	164.4	140.0	70.0	1000.0	16.4	14.0	7.0
Fe (mg)	6.2	6.2	1.8	14	44.3	44.3	4.1
Na (mg)	77.4	58.6	3.4	2000.0	3.9	2.9	0.2

F1 = Formulation 1 (70% of milk; 30% whey); F2 = Formulation 2 (50% of milk; 50% of whey); M = mangaba pulp; %DV = Daily Values are based on a diet of 2.000 kcal. RDI = Recommended daily intake

formulations, given that milk is the major calcium source. Mangaba also has substantial calcium contents (Silva et al. 2008). Calcium is essential to children development, due it is relationship with bone and tooth formation since this mineral supports organic tissue growth and development (FAO/WHO Expert Consultation 2005).

Sodium content was higher in F1 than in F2 (32%), this is explicated due milk naturally possesses high sodium contents. The reduction of sodium content in products is a goal for industries, once know about harmful effects associated to excessive sodium consumption. The iron supplementation corresponded to 44.3% of RDI (FAO/WHO Expert Consultation 2005), thus the products developed here can be considered as iron dietary sources. Monteiro et al. (2002) reported iron supplementation as a countermeasure for iron deficiency anemia in children, corroborating the benefits provided by the use of food products as iron carriers in combating this type of anemia.

Microbiological analysis

The presumptive test for total coliforms, which relied on the presence of gas and medium turbidity, was negative for both formulations. Thus, the confirmatory test in Brilliant Green Bile broth was not necessary. For yeasts and molds, no colonies were observed in both formulations after plate incubation. The formulations developed here met the criteria and minimal quality requirements for milk drinks intended for human consumption (BRASIL 2005a) and indicate the adoption of good sanitary condition during processing.

Sensory evaluation

From the 120 invited children and adolescents, 66 were allowed by their parents or legal guardians to participate in the survey, 61.0% and 39.0% of which were female and male, respectively, and 71.0% and 29.0% of which were

children (aging from 6 to 12 years) and adolescents (12 to 15 years), respectively. There were difference in acceptability between formulations and judge ages, according to ANOVA, $p = 0.0003$ and $p = 0.0001$, respectively, but the gender did not show effect about response.

The replacement of milk for whey increased the acceptability among judges, F2 showed acceptability of 85.0%, while F1 has not been approved by all judges. Regarding the age range of the judges, children showed highest acceptability than adolescents for both formulations. The average of acceptability to F1 was 78.7 and 5.2% for children and adolescents, respectively; while to F2, the acceptability was 91.5 and 73.6% for children and adolescents respectively.

As shown in Fig. 1, 78.7% of children judges answered “like” or “like extremely” of F1 fermented milk drink. However, only 19.0% of adolescents judges answered “like” of drink. To F2 fermented milk drink, the acceptability range 91.4% of children (like or like extremely) and 73.7% of adolescents judges answered “like” or “like extremely” (Fig. 2). The results achieved by F2 become the product suitable to include in school meal, once the acceptability level higher than 85% (BRASIL 2009).

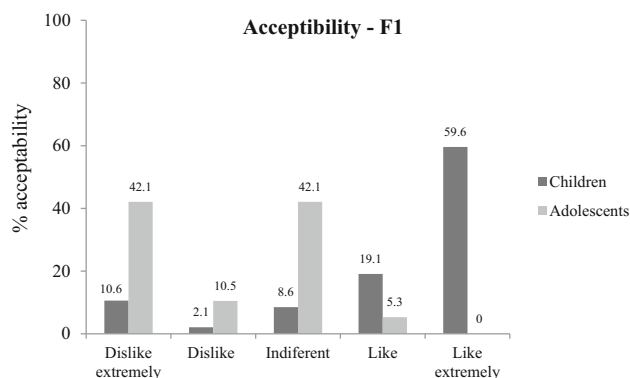


Fig. 1 Histogram of acceptance test of the fermented milk—F1 (70% of milk; 30% of reconstituted whey) by judges, considering ages

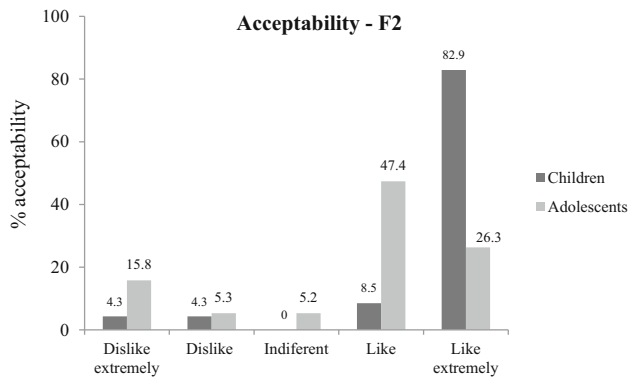


Fig. 2 Histogram of acceptance test of the fermented milk—F2 (50% of milk: 50% of reconstituted whey) by judges, considering ages

Figueiredo et al (2019) studied the acceptability of fermented milk drinks added by cerrado fruit pulp (cagaita, coquinho, tamarindo e umbu) and proved that the formulation containing 39% of whey had the highest acceptability. In this work, the judges attributed their choice to the pleasant texture, homogeneity, and flavor.

Some of the judges attributed the preference for F2 to the combination of mangaba's milder flavor and less thick texture, whereas F1 presented milk flavor and a thicker texture. The reason for this is the lowest whey concentration, showing a firmer clot. One of parameters that influence the acceptability is the texture, according to Koksoy and Kilic (2004) the stabilizer content to be added in dairy products influences their acceptability by consumers. Here, the stabilizer was added to avoid pulp water syneresis. Overall, flavor changes upon the addition of iron amino acid chelate were not perceived by the judges. Therefore, this product is recommended for dietary supplementation due to iron bioavailability.

Conclusions

The formulations developed showed contribution of protein and mineral aport. The use of whey in drink showed a great option to use of this cheaper raw and increased the acceptance of the product. The formulation of 50% of milk and 50% of whey was highly accepted by children (91.5%) adolescents (73.6%). In addition, the iron supplementation and mangaba pulp has a significant potential to improve of quality and preventing iron deficiency anemia. The formulations developed in this study are an excellent option to include in school feeding. Besides that, it is reported that an alternative means of taking advantage of the nutritional and technological properties of whey.

Acknowledgements This work was realized with the support of the FAPEMIG, CNPq, Ministério da Educação—Brasil (PROEXT), Pró-

Reitoria de Extensão da Universidade Federal de Minas Gerais (PROEX) and Pró-Reitoria de Pesquisa da Universidade Federal de Minas Gerais (PRPq).

Authors' contributions SMR: performed the experiments; SMR, BMAC, WJNL and CAFD analyzed data, development of milk drink with whey fermented and wrote the manuscript; SMR, CAFD, BMAC and IVB participated in the analysis and acceptability test by children and adolescents. GRLM, FAOR, GLM and IVB revised the manuscript for English language and technical aspects of the work.

Funding The funding for this Project was supported by Ministério da Educação—Brasil (PROEXT), Pró-Reitoria de Extensão da Universidade Federal de Minas Gerais (PROEX) and Pró-Reitoria de Pesquisa da Universidade Federal de Minas Gerais (PRPq).

Availability of data and material Not applicable.

Code availability Not applicable.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Consent to participate Informed consent was obtained from all individual participants included in the study.

Consent for publication Participants signed informed consent regarding publishing of results.

Ethics approval All experimental procedure with sensorial analysis were approved by the Ethics Committee at the Federal University of Minas Gerais (Process no 0465.0.203.000-10).

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