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Development And Evaluation Of A Vegetable Drink Based On Rice Milk, Quinoa And Passion Fruit Pulp

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Abstract:
A portion of the Brazilian population has an inability to digest, metabolize, or absorb food or some of its nutrients. Lactose intolerance is a disorder that occurs at all ages. Lactose-free products are a necessity to meet the needs of individuals who suffer from this disorder. Due to the demand for lactose-free foods, a plant-based beverage based on rice milk, quinoa, and flavored with passion fruit was developed as a nutritional beverage option. Formulations (F1, F2, and F3) were developed by varying the amounts of rice and quinoa and fixing the amounts of passion fruit pulp, water, and sucrose. The physicochemical analysis followed the physicochemical methods for food analysis of the Adolfo Lutz Institute and the microbiological analysis followed the Manual of Methods for Microbiological Analysis of Foods. In order to verify whether there were significant differences, the ANOVA analysis of variance was applied using the SISVAR program version 5.6. The Principal Component Analysis (PCA) was performed with the PAST software. The plant-based beverage made from rice milk, quinoa and passion fruit pulp had a low lipid index and low ash concentration, an acidic pH and 4.01% of the daily protein intake required for individuals aged 4 to 8 years. It was produced naturally, without stabilizers, thickeners or industrial emulsifiers. Its production could be a feasible technological possibility for small manufacturers. The rice and quinoa beverage flavored with passion fruit pulp could be a viable alternative for people seeking a balanced, nutritious diet or for people who are lactose intolerant or allergic to animal milk or soy proteins.
Key Word: *Lactose intolerance; Nutrition; Functional beverage; Food allergy.*

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I. Introduction

Children and adults suffer from nutritional problems such as allergies and food intolerances. It is estimated that 44% of the Brazilian population has some type of food intolerance [1]. These immunological disorders negatively impact the quality of life of those who suffer from them. Individuals with intolerance are unable to digest, metabolize, or absorb food or some of its nutrients. It is caused by the absence of a specific enzyme to process a certain nutrient [2]. Lactose is the disaccharide present in mammalian milk and its derivatives that is hydrolyzed by the enzyme lactase, producing two monosaccharides, glucose and galactose [3]. It is widely used in the production of various products such as mayonnaise, jelly, ice cream, and canned food [4].

Lactose intolerance is the difficulty in digesting or absorbing lactose due to the inability of the enzyme lactase to hydrolyze this disaccharide [5]. The receptivity of milk and its derivatives in organisms that have this enzyme deficiency can change depending on the level of their intolerance [6]. Lactose in absorption is a disorder that occurs at all ages, with primary intolerance being the most common in adults, presenting gas, diarrhea, abdominal pain and distension, and flatulence as symptoms [7]. Lactose-free products are a necessity to serve individuals who suffer from this disorder [9].

According to [10], it is necessary to develop methodologies to replace milk of animal origin with vegetable beverages extracted from oilseeds as infant food supplements for children with lactose intolerance. As an alternative to cow's milk, the lactose intolerant population has been using beverages extracted from legumes, oilseeds or cereals [11]. Vegetable milks, vegetable extracts or vegetable beverages are innovations developed by the food industry to meet the demand of individuals with restrictions on animal milk and its derivatives [12]. Vegetable beverages can be obtained from vegetable matrices, oilseeds, cereals, pseudocereals, legumes and fruits through maceration in aqueous solution, filtration, homogenization and heat treatment of the vegetable base with sensory, nutritional attributes and composition very different from milk of animal origin [13]. Rice and soy extracts are commercially viable [14].

Euromonitor International reports that 90% of vegetable beverages sold in the Brazilian market are soy-based [15]. Soybean vegetable extracts, widely used in the industry, have several occurrences of food allergies due to several proteins that cause this nutritional disorder [16] and unpleasant taste properties due to the enzymatic action of lipoxygenase, making it necessary to mix rice extract to make it palatable [17]. Rice extract has a sweet and mild flavor, starch and low protein, lipid, fiber and mineral content, which depending on the region, cultivation, type of grain and processing can vary in nutrient concentrations [18]. Several varieties of rice have a concentration of phenolic compounds ranging from 72.45mg to 120.13mg and high antioxidant activity [19].

Rice milk is considered a healthy drink that is related to several advantages that benefit the consumer's health and its nutritional composition [20]. It is an alternative to cow's and soy milk for individuals with lactose intolerance and allergies [21]. Rice milk is a sweet-tasting and highly digestible drink with low production costs, requiring lipid and protein supplements to be used as food for children [22]. The minerals iron, zinc, calcium and magnesium, found in rice, have greater bioavailability, being absorbed more than those found in animal milk [23].

Quinoa is a monocotyledonous pseudo-cereal of Andean origin that does not belong to the cereals (dicotyledons), but they are nutritionally similar. [24]. It is a grain that is little consumed by most Brazilians who are unaware of its nutritional advantages. It has considerable amounts of proteins, vitamins, lipids, essential amino acids, fiber, flavonoids, polyphenols, and phytosterols that provide numerous health benefits [25]. Quinoa is also rich in phosphorus and vitamins E and B complex, and has higher levels of iron and calcium than corn, rice, oats and wheat [26]. Quinoa has a high percentage of protein compared to traditional cereals and high nutritional value [27]. It can be included in the diet of people with celiac disease, aiding in child development and preventing chronic diseases due to the presence of the essential amino acid histamine [28]. The water-soluble extract of quinoa must be produced by immersing the grains in water for a long period, then grinding, filtering and homogenizing [29].

In view of the demand for lactose-free and allergy-causing foods, which are diverse and unavailable in the consumer market, a plant-based drink based on rice milk supplemented with quinoa and flavored with passion fruit was developed as a nutritional drink option.

II. Material And Methods

The rice milk was extracted, the quinoa processed and the vegetable drink developed in the Industrial Kitchen and Food Laboratory of the Federal Institute of Education, Science and Technology of Tocantins - IFTO: Paraíso do Tocantins campus. The ingredients - passion fruit pulp, agar gum, rice in natura, quinoa and probiotic milk culture - used in the processing, extraction and preparation were purchased from local businesses in the city of Paraíso do Tocantins.

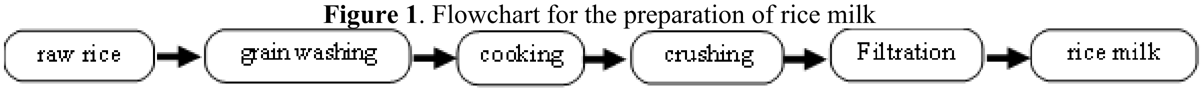
Hygienization of utensils

All the utensils used were sanitized and disinfected in boiling water for 20 minutes.

Making the rice milk

Rice milk was prepared according to the methodology adapted from [30], using fresh rice and boiling water. Flowchart 1 shows the steps involved in preparing rice milk.

Figure 1. Flowchart for the preparation of rice milk



Source: Authors, 2024

Preparation of the pulp

Passion fruit pulp was purchased in 100-gram packages, within the expiry date, from the shops in the city of Paraíso do Tocantins. The pulp samples were packed in thermal boxes at a temperature of 4 °C and transported immediately to the Industrial Kitchen of the IFTO Paraíso do Tocantins campus, where they will remain frozen at -18 °C until they are used in the process of making the vegetable drink. The fruit pulps were thawed at room temperature, 25 °C, homogenized in an industrial blender and added to the vegetable drink produced.

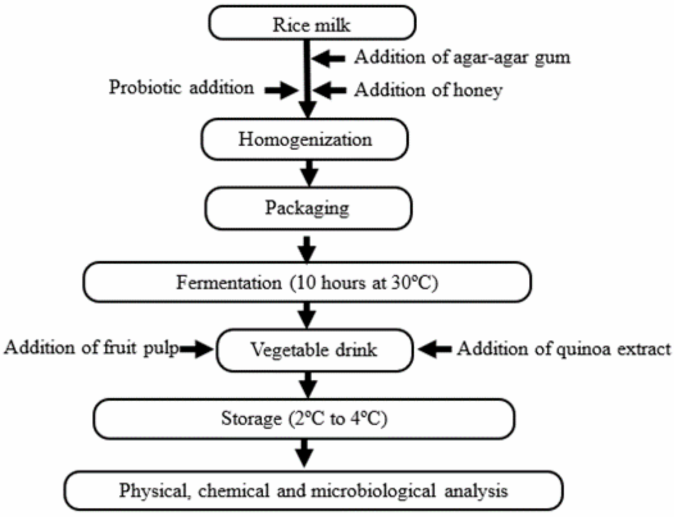
Preparation of the quinoa extract.

The extracts were prepared according to the adapted methodology proposed [10]. The grains were boiled for 20 minutes in a 0.25% sodium bicarbonate solution. After cooking, the grains were washed and disintegrated with water at boiling temperature using an industrial disintegrator. The resulting dispersion was filtered and stored under refrigeration at 4 °C in glass jars with lids until it was incorporated into the fermented milk produced.

Production of vegetable drink with rice milk, quinoa and fruit.

The production of the vegetable drink with rice milk used the methodology adapted from [30]. Rice milk, agar-agar gum and the probiotics Lactobacillus acidophilus, Bifidobacterium and Streptococcus thermophilus were used to make the drink. Flowchart 2 shows the production stages of the vegetable drink.

Figure 2. Stages in the production of vegetable drinks with rice milk, quinoa and fruit.



Source: Adapted from Brandão *et al*., 2021

Vegetable drink formulations

Three (3) formulations were made (F1, F2 and F3) varying the amounts of rice and quinoa. Table 1 shows the proportions of the ingredients used for the formulations evaluated in this study.

Table 1. Variations in the proportions of rice and quinoa in the vegetable drink produced.

Ingredients (%)	FBV1	FBVF2	FBV3
Rice extract (%)	20	15	10
Quinoa extract (%)	10	15	20
Fruit pulp (%)	20	20	20
Sucrose (%)	20	20	20
Water (%)	30	30	30

Source: Authors, 2024

To prepare the drink, the rice and quinoa extracts and the other ingredients were blended in a blender for 5 minutes. The drink was pasteurized at 75 °C for 15 minutes, filled into glass jars with metal lids, previously sterilized in an autoclave at 121 °C for 30 minutes, and kept refrigerated at a temperature of 4 ± 1 °C.

Physical-chemical analysis.

The physicochemical characteristics:
Carbohydrates: by oxidation-reduction titration [31];
Ash by the waste incineration method [31];
Total fats by the Soxhlet method [31];
Hydrogen potential (pH): direct reading on a digital potentiometer [31];
Proteins by the Kdjedahl method [31];
Moisture by the oven drying method at 105 °C for 4 h [31];

were carried out in triplicate at the Food Analysis Laboratory of the IFTO: Paraíso do Tocantins campus, following the physicochemical methods for food analysis of the Adolfo Lutz Institute [31] and the Official Methods of Analysis of the Analytical Chemistry Association [32].

Microbiological analysis.
Determinations of coliforms at 45 °C (*Escherichia coli*) and *Salmonella* sp. were carried out according to the methodology of [33] and the results were compared with the Normative Instruction - IN no. 161, of July 1, 2022 of the National Health Surveillance Agency - ANVISA [34].

Statistical analysis

In order to check whether there was a significant difference between the results, ANOVA analysis of variance was applied and Tukey's test was applied between the means of the response variables at a 5% significance level, using the SISVAR program version 5.6 [35]. Principal Component Analysis (PCA) assessed the interrelationship between the data and the treatments according to similarity. The PCA analysis was carried out using the PAST software [36].

III. Results And Discussion

The results of the physical-chemical parameters total carbohydrates, ash, lipids, hydrogenionic potential (pH), proteins, total soluble solids and moisture analyzed in the three drink formulations with different proportions of rice and quinoa extract are shown in Table 2.

Table 2. Physico-chemical analysis of the vegetable drink formulations (FBV) made from rice milk and quinoa.

Physical-chemical parameters	FBV1	FBV2	FBV3
Total carbohydrates (%)	$11,638^{C} \pm 0,100$	$11,375^{B} \pm 0,220$	$11,356^{A} \pm 0,100$
Ash (%)	$0,360^{A} \pm 0,010$	$0,440^{A} \pm 0,010$	$0,560^{A} \pm 0,020$
Lipids (%)	$0,390^{A} \pm 0,010$	$0,460^{B} \pm 0,100$	$0,520^{C} \pm 0,010$
pH	$3,890^{A} \pm 0,380$	$3,933^{B} \pm 0,240$	$4,026^{C} \pm 0,430$
Proteins (%)	$0,705^{A} \pm 0,000$	$0,766^{B} \pm 0,060$	$0,816^{C} \pm 0,070$
Moisture (%)	$87,532^{A} \pm 0,090$	$87,719^{A} \pm 0,210$	$87,839^{A} \pm 0,120$

Different letters on the same line indicate a significant difference ($p < 0.05$)
Source: Authors, 2024

There were significant differences ($p < 0.05$) between the treatments analyzed. These differences may be associated with an increase in the amount and capacity of quinoa grains to absorb water during the hydration time. A study carried out by [37] showed that 100 grams of quinoa absorbed 71 grams of water in 2 hours of hydration.

The FBV1 formulation had a higher percentage of carbohydrates than FVB2 and FVB3, as carbohydrates are the main constituent of the rice extract in these formulations. Research carried out by [38], who developed and characterized a chocolate drink based on water-soluble extracts of quinoa and rice, obtained carbohydrate contents of 16.58% ± 0.042, 13.77% ± 0.194 and 14.33% ± 0.055 respectively for the three formulations produced.

With regard to the physicochemical analysis of ash and moisture, the three formulations did not differ significantly ($p > 0.05$). The ash and moisture contents found in this research are, respectively, above and below the values found by [39]. The research also showed higher ash and moisture values than those found by [38].

In terms of lipids, the FBV3 formulation showed the highest percentage. The progressive increase in quinoa extract may have contributed to the increase in lipid content in the FBV3 formulation. The lipid content found by [40] when preparing and characterizing a fermented drink based on water-soluble quinoa extract with fruit pulp, was higher than that found in this research.

The FBV3 formulation showed the highest value for hydrogen potential. The water absorbed by the quinoa during the hydration process may have contributed to a reduction in acidity and consequently an increase in hydrogen potential. A pH above 4.5 in vacuum-stored drinks can lead to the development of *Clostridium botulinum* [41].

Among the three formulations presented, FBV3 had the highest protein content. The average protein content for FBV1, FBV2 and FBV3 is 0.762%. According to [42] and [43], protein intake can be calculated taking 19 grams as 100% of the daily requirement, so 100 grams of the drink produced in this study provided 4.01% of the daily protein requirement for individuals aged between 4 and 8 years.

The results of the microbiological analysis of the vegetable drink based on rice milk, quinoa and passion fruit pulp are shown in Table 3.

Table 3. Results of the microbiological analysis of vegetable drinks based on rice milk, quinoa and passion fruit pulp.

Microorganisms	FBV1	FBV2	FBV3
Escherichia coli ($NMP.mL^{-1}$)	< 3	< 3	< 3
Salmonella spp. (25 mL)	Absence	Absence	Absence

Source: Authors, 2024

Due to the lack of Brazilian legislation for vegetable drinks based on rice milk, quinoa and passion fruit pulp, the results were compared with the microbiological parameters of juices and other “in natura” or reconstituted drinks recommended in IN No. 161/2022 [34]. According to the values shown in Table 3, the vegetable drink based on rice milk, quinoa and passion fruit pulp complied with the food safety and consumption conditions required by the legislation and posed no risk to consumer health. Carrying out microbiological research on chocolate vegetable drinks, [38] also found a result that complied with the legislation and posed no risk to consumer health. The good manufacturing practices used in the production of the vegetable drink contributed to safety and the absence of microbiological contamination.

Principal component analysis (PCA) was applied to better interpret the data and observe similarities or differences between the FBV1, FBV2 and FBV3 formulations based on their physicochemical characteristics, as shown in Table 2.

In this way, PCA projected the data into two more significant principal components with their respective eigenvalues, explained variance and accumulated variance (table 4).

Table 04. Principal components (PC), eigenvalues (eigenvalue) and percentage of explained variance

PC	Eigenvalue	Variance explained
1	5,749	95,823%
2	0,251	4,177%

Source: Authors, 2024

Based on the results obtained by principal component analysis, the respective eigenvalues and percentages of variance, shown in Table 4, indicate that PC1 was responsible for 95.823% and PC2 for 4.177% of the variations in the data. The sum of principal components I and II adequately presented the variability between the samples [44].

The weights of the principal components CP1 and CP2 for the physical-chemical variables total carbohydrates, ash, lipids, pH, proteins and moisture are shown in Table 5.

Table 5 - Highest values in bold of the components for the variables analyzed

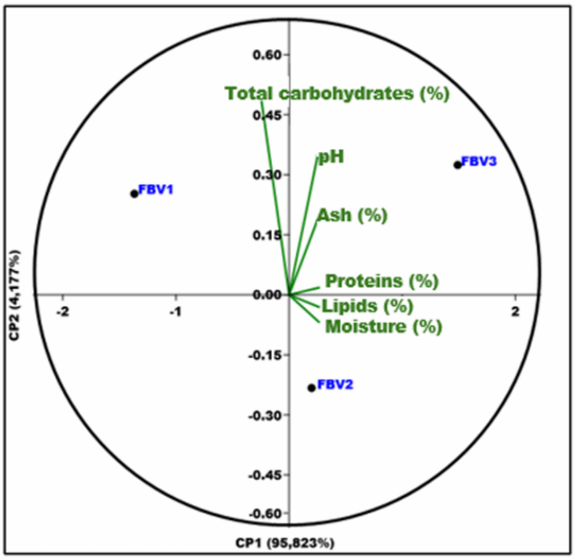
Physical-chemical parameters	CP1	CP2
Total carbohydrates (%)	**0.41208**	0.30753
Ash (%)	-0.38521	**0.76551**
Lipids (%)	**0.41693**	-0.048126
pH	**0.41701**	0.028106
Proteins (%)	0.40081	**0.55205**
Moisture (%)	**0.41645**	-0.10749

Source: Authors, 2024

According to the data shown in Table 5, the first principal component (CP1) has greater weight for the variables total carbohydrates (%), lipids (%), pH and Moisture (%) and the principal component (CP2) has greater weight for the variables ash (%) and protein (%).

Graph 01 shows the main components (CP1) and (CP2), displaying their respective weights, making it possible to visualize the main groupings in the set of variables.

Figure 01. Graph of the principal component analysis for the collection points and physical-chemical parameters.



Source: Authors, 2024

CP measurements are interpreted according to the correlations between each parameter and each principal component, so measurements close to each other are positively correlated, measurements 180° apart are negatively correlated, while if they are 90° apart they are independent.

Graph 1 (Figure 01) shows that the formulations FBV2 and FBV3 are related to the main component CP1 and FBV1 to the main component CP2. pH, ash, and proteins had a great influence on the characterization of CP2, being negatively correlated with total carbohydrates, lipids, and moisture.

The FBV1 formulation had a higher carbohydrate content and a lower ash, lipid, pH, protein and moisture content than the FBV3 formulation. The FBV2 formulation showed intermediate percentage values between FBV1 and FBV3. The formulation FBV3 had the highest levels of ash, lipids, pH, protein and moisture.

IV. Conclusion

The plant-based drink made from rice milk, quinoa and passion fruit pulp demonstrated low lipid index and low ash concentration, acidic pH, excess moisture and 4.01% of the daily protein intake required for individuals aged 4 to 8 years. It is produced naturally, without stabilizers, thickeners and industrial emulsifiers. Its production may be an executable technological possibility for small manufacturers. The rice and quinoa drink flavored with passion fruit pulp may be a viable alternative for people seeking a nutritious, balanced diet, who are lactose intolerant or allergic to animal milk or soy proteins.

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