

Drink with probiotic potential based on water-soluble extract from cashew nuts

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ABSTRACT: The research elaborat a drink based on a water-soluble cashew nut extract (WCNE), fermented with the probiotic strain Lactobacillus paracasei ATCC 334. The formulations were defined by factorial design 2^2 , with the independent variables: concentration of Lb. paracasei (1%, 1.5%, and 2%) and cashew nuts (100 g, 200 g, and 300 g), and the dependent variables: Composition (Moisture, Lipids, Proteins, Ashes and Carbohydrates) and Sensory Analysis (acceptance and ordination tests). The Lb. paracasei concentration in the WCNE registered between 7.72 log CFU g⁻¹ to 8.27 log CFU g⁻¹ for 6 hours of fermentation. According to the FDA, these values indicated a probiotic potential to the formulations (above 6 log CFU g⁻¹). The concentration of cashew nuts (CNC) had a significant influence (P < 0.05) on all physical-chemical parameters and sensory attributes of the fermented WCNE, demonstrating itself as a determining factor for the product nutritional quality. The concentration of Lb. paracasei affected significantly (P < 0.05) the reduction of humidity and the increase of lipids and sh contents. The drinks F3 (300g CNC, 1% Lb. paracasei) and F4 (300g CNC, 2% Lb. paracasei) obtained the highest scores in the acceptance test and in the attributes of aroma, flavor, and overall impression. The study indicated that, in addition to good sensory acceptance, fermented WCNE can provide a concentration of probiotics above 7 log CFU g-1, thus being a promising alternative to meet the daily demand of probiotics for people who are restricted to dairy products.

Key words: Anacardium occidentale L. Functional Foods. Lactobacillus paracasei, Vegetable Extract.

Bebida com potencial probiótico à base de extrato hidrossolúvel de castanha de caju

RESUMO: A pesquisa teve como objetivo a elaboração de uma bebida à base de extrato hidrossolúvel de castanha de caju (EHCC), fermentada com a cepa probiótica Lactobacillus paracasei ATCC 334. As formulações foram definidas pelo planejamento fatorial 2^2 , com as variáveis independentes: concentração de Lb. paracasei (1%, 1,5% e 2%) e castanha de caju (100 g, 200 g e 300 g), e as variáveis dependentes: Composição (Umidade, Lipídios, Proteínas, Cinzas e Carboidratos) e Análise Sensorial (testes de aceitação e ordenação). A concentração de Lb. paracasei no EHCC apresentou valores entre 7,72 log UFC/g a 8,27 log UFC/g com seis horas de fermentação. De acordo com o FDA, esses valores indicam o potencial probiótico das formulações (acima de 6 log UFC/g); a concentração de castanha de caju (CCC) teve influência significativa (P < 0,05) em todos os parâmetros físico-químicos e atributos sensoriais do EHCC fermentado demonstrandose como fator determinante para qualidade nutricional do produto; a concentração de Lb. paracasei afetou significativamente (P < 0,05) a redução da umidade e o aumento dos conteúdos de lipídios e cinzas. As bebidas F3 (300 g CCC, 1% Lb. paracasei) e F4 (300 g CCC, 2% Lb. paracasei) obtiveram as maiores notes te de aceitação e nos atributos aroma, sabor e impressão global. O estudo indicou que, além da boa aceitação sensorial o EHCC fermentado pode fornecer concentração de probióticos acima de 7 log UFC g⁻, sendo assim uma promissora alternativa para atender a demanda diária de probióticos à pessoas que apresentam restrição à produtos lácteos. **Palavras-chave:** Alimentos funcionais. Anacadium occidentale L. Extrato vegetais. Lactobacillus paracasei.

INTRODUCTION

The *Lactobacillus paracasei* are considered probiotic microorganisms responsible for improving or maintaining the microbial balance in the intestine, assisting in vitamin production, promoting gastrointestinal and urogenital resistance against

pathogens. They also increase lactose digestibility in lactose-intolerant people. They confer benefits to hosts' health when administered in adequate amounts (BRASIL, 2018; GROM et al., 2020).

To be considered with probiotic potential, a product must contain viable, specific microorganisms in sufficient concentration able to alter the hosts'

Received 03.18.21 Approved 10.12.21 Returned by the author 05.22.22 CR-2021-0218.R2 Editors: Rudi Weiblen D Cristiano Ragagnin de Menezes D microflora by implantation or colonization and promoting beneficial effects on host health (ZIELIŃSKA; KOLOŻYN-KRAJEWSKA, 2018.). The classification as probiotic to microbial species requires the characterization of the microorganisms used in their production, their genetic profiles of resistance to antimicrobials, case reports on possible adverse effects of the strains, demonstration of their effectiveness and viability (BRASIL, 2018).

The increase in adherents to vegetarianism and veganism drives the food industry to diversify. This scenario also motivates the dairy industry to embrace the development of new products, such as "alternative milks", which elevates the variety of probiotic products based on vegetables. This diversification of products also favors consumers who are lactose intolerant and allergic to milk protein, since 75% of the world population suffers from some degree of lactose intolerance, increasing the demand for alternative products to milk (PANGHAL et al., 2017;).

Cashew nuts represent an option for the manufacture of water-soluble extracts of vegetable origin. These nuts provide large amounts of energy, with an average of 596 kcal 100 g⁻¹, and fat (46%), primarily composed of essential fatty acids, such as linoleic and oleic acids, which act in free radical reduction and cardiovascular diseases control (MOHAN et al., 2018).

The national production of cashew nuts occupies a prominent place in Brazil, raising annual values of US\$ 100 million, generating economic and social activity in contry's Northeast region, which concentrates 99.7% of Brazilian plantations. Cashew exportation corresponds to about 90% of national production (BRAINER & VIDAL, 2020). However, the fruit processing results in a higher percentage of broken nuts. This fact reflects their price depreciation, if compared to whole ones. The processing of these broken nuts for water-soluble extract formulation presents itself as an alternative for reuse. Therefore, the present research proposed to elaborate a fermented drink from the water-soluble cashew nut extract added with the probiotic Lactobacillus paracasei, representing a viable, nutritious product with functional potential and an option for people with dietary restrictions.

MATERIALS AND METHODS

Cashew nuts and probiotic culture aquisitions

The cashew nut (*Anacardium occidentale* L.) came from local stores in the Metropolitan Region of Recife. The experiment mother

culture, *Lactobacillus paracasei* ATCC 334, in the concentration of 13 log CFU g⁻¹, was provided from the culture collection of the Food and Environment Laboratory of the Rural Technology Department of the Federal Rural University of Pernambuco - UFRPE. The concentrate was created by producing biomass from the *Lb. paracasei* culture donated by the laboratory, using Agar MRS (Man Rogosa & Sharpe) by theslanted tubes technique, incubated in anaerobic conditions for 48h at 35 °C. After this period, the cultures were washed with a sterile solution containing water- soluble extract of cashew nut (at 10% of cashew nut per liter of water) with 20% glycerol, and stored at -18 °C for later use. The experiments were carried out in triplicate.

Preparation of the WCNE

The cashew nut selection followed the criteria of absence of dark spots on nuts surface, presenting a uniform white color. They were washed and sterilized (121 °C 15 min⁻¹). After this stage, the nuts were submerged in sterile distilled water for overnight hydration, under refrigeration (4 $^{\circ}C \pm 2$), following a nuts: water ratio of 1: 2. After the nuts swelling, the excessive water was drained in aseptic conditions, and the cashew nuts were processed, producing the water-soluble cashew nut extract (WCNE). The hydrated cashew nuts were crushed with sterile distilled water in a domestic blender (Mondial, Power 2i NL-26), for 5 minutes, in the concentrations of 100 g (C1), 200 g (C2), and 300 g (C3) of cashew nuts to 1 L of sterilized water. WCNE went through a 35 mesh (0.5 mm) sieve, bottled in sterile flasks with screw caps, and were pasteurized $(72 \text{ °C} \pm 2 \text{ C}^{\circ} 20 \text{ min}^{-1})$, followed by immersion in an ice bath for 20 minutes (MORAIS et al, 2010). The experiments were carried out in triplicate.

Experimental design of fermentation

pasteurization, WCNE After the formulations were inoculated with the mother culture at a concentration of 13 log CFU g⁻¹, following an experimental design defined by factorial design 2², which intended to evaluate the influence of nut and probiotic concentrations over the parameters of nutritional composition and sensorial attributes by the surface response methodology. The independent variables were concentration of Lb. paracasei (1%, 1.5%, and 2%) and cashew nuts (100 g, 200 g, and 300 g). They were combined producing 4 factorial points and 3 repetitions of the central point. Time and temperature of fermentation were constant through every combination. The experiments were carried out in triplicate.

After the inoculation, the extracts were incubated at 35 °C, and fermented for 6 hours. Then, the WCNE was stored under refrigeration at 4 °C \pm 2 for posterior analysis (TABANELLI et al., 2018).

Microbiological analysis

Previously to the probiotic inoculation, the pasteurized extracts were submitted to microbiological analysis. The quantification of total and thermotolerant coliforms was performed by the multiple tube technique, expressed in Most Probable Number per gram (NMP g⁻¹) (APHA, 2001). The presence/absence of *Salmonella* sp., total mesophiles, and psychotropic counts used commercial Compact Dry[®] kits, all approved by Codex Alimentarius, ICMSF, APHA, FDA, ISSO Standards, AOAC. The experiments were carried out in triplicate.

Viability of Lb.paracasei

The viability test of *Lb.paracasei* ATCC 334 in the WCNE was quantified by the pour plate method, using serial dilutions up to 10^{13} , using MRS Agar (Man Rogosa & Sharpe), incubated in anaerobiosis for 48h at 35 °C \pm 2. All analysis were performed in triplicate. The count was expressed in Colony Forming Unit per gram (CFU g⁻¹) (SILVA et al., 2017).

Physical chemical analysis

The centesimal composition of WCNE formulations involved the following quantifications: proteins by the Kjeldahl method, with a nitrogen correction factor of 5.46; ash content by incineration (550 C°); lipids by Soxhlet; moisture by the infrared drying method, using an Infrared scale (ID50 Marconi), according to the Association of Official Analytical Chemistry (AOAC, 2016). The total carbohydrate value was calculated by difference. The experiments were carried out in triplicate.

The pH was determined by the electrometric method in a potentiometer, using 5 mL of each sample diluted in 10 mL of distilled water. Subsequently, the same samples went through total titratable acidity determination, with NaOH (0.1N), using phenolphthalein (1% w/v) as a turning indicator. The results expressed in oleic acid (AOAC, 2016). The experiments were carried out in triplicate.

Sensory analysis

The research was previously submitted for approval by the Ethics Committee of the University of Pernambuco (CAAE: 25725019.4.0000.5207). The sensory analysis took place at the Laboratory of Sensory Analysis of Gastronomy at the Department of Rural Technology (DTR/UFRPE). The panel was formed by 100 individuals of both sexes, untrained, between 18 and 60 years-old. The fermented WCNE formulations were sweetened with Stevia powder for culinary use (Stevita Forno e Fogão), in the proportion of 10g for each 1 liter of drink (REBOUCAS et al., 2014).

The panelists received five samples for both tests. The acceptance test was performed for the attributes: aroma, appearance, flavor, and global impression, using a structured hedonic scale of nine points (1 = extremely disliked; 9 = extremely liked). The ordination test was also applied to classify the samples according to preferences in ascending order of color, flavor, texture, and odor. Each panelist also received mineral water at room temperature and salt crackers to rinse the papillae, a consenting form, and forms for both tests. The samples served in both tests contained 30 ml of each formulation, refrigerated (5 \pm 1°C), and randomly coded (IAL, 2008).

Statistical analysis

The experiments were carried out in triplicate, for calculation of means and standard deviation. The results for physical-chemical and sensory analysis were evaluated through ANOVA (analysis of variance) using the Duncan test (P < 0,05) for comparison of means through the program "Statistic for Windows 5.0".

RESULTS AND DISCUSSION

WCNE Microbiological analysis

formulations All were absent from Salmonella sp. in 25 milliliters. The pasteurized WCNE formulations were within legal limits since the concentration for total mesophiles and psychotropics were 20 CFU mL⁻¹ (0.30 Log of CFU mL⁻¹) and <10 CFU mL⁻¹, respectively. Superior values were detected in the microbiological analysis of a watersoluble pasteurized extract from Tiger nut (Cyperus esculentus L.) by Codina-Torrella et al. (2018), who reported 5.51 Log CFU mL-1 and 5.37 log CFU mL-1 of psychotropics and aerobic mesophiles, respectively. These microorganisms are responsible for a rapid deterioration and alteration of these products through the fermentation of sugars, production of acid and gas, resulting in shelf-life reduction.

The legal basis for WCNE similar products is the MAPA Normative Instruction N^o 19 (MAPA, 2013), which indicates the standard of identity and quality for ready-to-eat drinks based on vegetables. All formulations presented a count <3.0 NMP mL⁻¹ for thermotolerant coliforms, indicating that the

sterilization of cashew nuts and the pasteurization of WCNE production were effective measures for microbiological control.

Count of Lactobacillus paracasei ATCC 334, pH and Titratable acidity.

The concentration of probiotics can be lowered, in comparison to its initial concentration, due to adaptation to a new substrate. Thus, time and temperature of fermentation are required to reach the logarithmic phase and increase the cells *Lb. paracasei*, the primary metabolites production, such as organic acids, and promoting a pH reduction. Table 1 shows the pH values, titratable acidity, and the concentration of *Lb. paracasei* from the WCNE formulations at the beginning and after 6 hours of fermentation.

All formulations showed probiotic amounts superior to 6 log CFU g⁻¹ (Table 1), with F2 showing the highest growth (8.27 log CFU g⁻¹) in just 6 hours of fermentation. Higher concentrations of probiotics in foods are required so that these microorganisms pass through the gastrointestinal tract and reach the proliferation capacity in the host intestine. Although there is no universal consensus, values ranging from 6 log CFU g⁻¹ to 8 log CFU g⁻¹ are considered acceptable in several countries. The recommendation of the Food and Drug Administration (FDA, 2003) is that in foods with probiotic function, their concentrations should be at least 6 log CFU g^{-1} (NOROUZI et al., 2019; PANGHAL et al., 2017; SANTOS et al., 2020).

The *Lb. paracasei* grows satisfactorily in plant substrates, as reported by Santos et al. (2020). Cashew nuts contain several carbohydrates such as glucose, maltose, lactose, fructose, cellobiose, raffinose, and stachyose. It is also a source of essential amino acids, such as arginine, vitamins (E, K, B6, and C), and minerals such as iron, phosphorus, magnesium, calcium, potassium, and selenium (MOHAN et al., 2018). These nutrients can be utilized as substrates for *Lb. paracasei* growth, due to its heterofermentative character, which corroborates with other researches that applied vegetable raw materials for probiotic drinks production (NOUROZI et al. 2019).

Time plays a key role in the growth of probiotics, since the fermentation process is associated with a reduction in pH. In the production of fermented milk and traditional yogurts the pH can reach 4.5, reaching viability of cultivation between 6 log UFC g⁻¹ and 7 log UFC g⁻¹ (GU et al., 2020). Although the lowest pH value obtained did not surpass 5.70, the formulations showed good cell growth in a few hours of fermentation. For Tabanelli et al. (2018), long-term fermentations of cashew nuts can lead to the occurrence of unwanted flavors, increasing the probability of sensory rejection of the product.

F	Concentration of <i>Lb.</i> <i>paracasei</i> (%)	Concentration of cashew nuts (g)	ILC (log CFU g ⁻¹)	FLC (log CFU g ⁻¹)	Initial pH	Final pH	IATT(g/100g)	FATT(g/100g)
F1	1(-1)	100 (-1)	7.36± 0.13	$8.23{\pm}0.12$	6.94± 0.02	5.88± 0.01	$0.776{\pm}\ 0.05$	$1.024{\pm}~0.08$
F2	2(1)	100(-1)	7.51 ± 0.18	$8.27{\pm}~0.15$	6.89± 0.01	5.70± 0.01	$0.769{\pm}0.05$	1.152 ± 0.09
F3	1(-1)	300(1)	7.04 ± 0.22	7.72 ± 0.31	$\substack{6.22\pm\\0.01}$	5.97± 0.03	$0.785{\pm}0.09$	0.896 ± 0.06
F4	2(1)	300(1)	7.07 ± 0.12	$8.11{\pm}0.17$	6.18± 0.05	5.95± 0.01	$0.791{\pm}0.04$	0.960 ± 0.08
F5	1,5(0)	200(0)	$7.27{\pm}~0.20$	$8.16{\pm}0.11$	6.19± 0.01	5.93± 0.02	$0.792{\pm}\ 0.08$	1.088 ± 0.01
F6	1,5(0)	200(0)	$7.27{\pm}~0.20$	8.16 ± 0.11	6.19± 0.01	5.93± 0.02	$0.792{\pm}\ 0.08$	1.088 ± 0.01
F7	1,5(0)	200(0)	$7.27{\pm}0.20$	8.16± 0.11	6.19± 0.01	5.93± 0.02	$0.792{\pm}0.08$	$1.088{\pm}~0.01$

Table 1 - Matrix planning for the fermentation of water-soluble cashew nut extract, according to the factorial variables and extract aspects.

*Values expressed as mean and standard deviation. F = Formulation. ILC = initial *Lb. paracasei* concentration. FLC = final *Lb. paracasei* concentration. ICC: initial cell concentration (0 hours). FCC: final cell concentration (6 hours). IATT: initial total titratable acidity. FATT: final total titratable acidity.

Nutritional composition of water-soluble cashew nuts extracts - pasteurized and fermented with *Lb. paracasei* ATCC 334.

Table 2 shows the results of factorial planning 2^2 for the nutritional parameters of fermented WCNE formulations. Table 3 presents the effects of the independent variables: *Lb. paracasei* concentration (%) and nut concentration (%) on the physical-chemical parameters.

According to table 3, only the protein content obtained a satisfactory response, with a non-significant lack of fit (P < 0.05). Other results depended on the two independent variables surveyed. The concentration of cashew nut was the one that obtained the greatest significant influence (P < 0.05) in all physical-chemical parameter, directly related to the availability of nutrients present in the extract.

The moisture parameter was contrary to other components. A lower concentration of cashew nuts was associated with higher moisture (Figure 1A), as presented in F1 (91.09%) and F2 (92.76%), which have the highest moisture values and lowest concentrations of cashew nuts. The other formulations varied between 84.01% - 90.06% (Table 2). The high moisture of WCNE is justified considering that it is a colloidal suspension consisted mainly of water (KEHINDI et al., 2020; PANGHAL et al., 2017).

F3 (9.09%) and F4 (6.64%) presented the highest lipids contents (Table 2). These values indicate that the fermented WCNE is rich in fat, as required in RDC n° 54/2012 of ANVISA (BRASIL, 2012). Figure 1 (B - E) displays that with the increase in the percentage of cashew nuts, the concentrations of lipids, proteins, carbohydrates, and ashes were higher, resulting in the addition of nutritional value. This behavior corroborates the results in Table 2 since F3 and F4 presented higher values of these parameters due to their higher proportions of cashew nuts. The nutrients concentrations in water-soluble plant extracts are directly related to the raw material proportions, whether the product is fermented or not (KEHINDI et al., 2020). The concentration of *Lb. paracasei* influenced moisture, lipids, and ashes (Table 3), unlike proteins and carbohydrates (P < 0.05), as can be seen in figure 1 (C and D).

The high-fat concentration in nuts, such as cashew nuts, may promote lipolysis compounds accumulation, presenting free fatty acids (FFA) after fermentation. TABANELLI et al. (2018) observed a significant increase (P < 0.05) in monounsaturated fatty acids (MUFA) and presence of free fatty acids (FFA) after fermentation of the cashew nut fermented cream, compared to raw nuts. The oleic acid present in cashew nuts is classified as MUFA and composes the omega-9 family, which are significant allies in the prevention of several types of cancers and reduction of free radicals, enabling the reduction of oxidative damage to DNA (MOHAN et al., 2018).

Ashes represent the mineral matter in cashew nuts. The most abundant minerals in cashew nuts are phosphorus, potassium, magnesium, followed by calcium, iron, copper, manganese, zinc, and selenium (AMORIM et al., 2018). The fermented WCNE showed ash concentrations between 0.06% to 0.27% (Table 2). Rebouças et al. (2014) obtained 0.31% of ash in the mixed drink of passion fruit

Table 2 - Centesimal composition from fermented WCNE and Average scores of sensory attributes.

Formulation	Moisture (%)*	Lipids (%)*	Proteins (%)*	Carbohydrates (%) [*]	Ashes (%)*	Aroma*	Appearance*	Flavor*	Global Impression *
F1	91.09 ^b	5.52°	1.92°	1.42°	0.06°	5.62ª	5.59 ^b	4.63 ^b	4.98 ^b
F2	92.76ª	4.08 ^c	1.52 ^d	1.57°	0.07°	5.71ª	5.72 ^b	4.40 ^b	5.02 ^b
F3	84.01°	9.09 ^a	3.51ª	3.20ª	0.19 ^b	6.19ª	6.30ª	5.63ª	5.93ª
F4	86.15 ^d	6.94 ^b	3.28ª	3.35ª	0.27^{a}	6.07 ^a	6.08 ^{ab}	5.45ª	5.81ª
F5	90.04°	4.87 ^d	2.98 ^b	2.99 ^b	0.17^{b}	5.65ª	6.22ª	5.32ª	5.52 ^{ab}
F6	90.06°	4.90 ^d	2.53 ^b	2.92 ^b	0.18 ^b	5.70ª	6.20 ^a	5.40 ^a	5.60 ^{ab}
F7	90.00°	4.86 ^d	2.45 ^b	2.93 ^b	0.17 ^b	5.60ª	6.23ª	5.37ª	5.50 ^{ab}

*Means followed by equal letters in the vertical do not differ at the level of 5% by Duncan's test. WCNE = water-soluble cashew nut extract. F = formulation.

0.04

0.954

NS

0.674

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Independent variables	Moisture (%)	Lipids (%)	Proteins (%)	Carbohydrate s (%)	Ashes (%)	Aroma	Appearance	Flavor
Concentratio n of <i>Lb.</i> <i>paracasei</i> (%)	1.90	-1.79	NS	NS	0.05	NS	NS	-0.20
Concentratio	-6.84	3 21	1.67	1 78	0.16	0.46	0.53	1.02

NS

0.854

Table 3 - Effects of Lb. paracasei and Cashew nut Concentrations (%) on physicochemical parameters and sensory attributes.

Lack of

1 e 2

 \mathbb{R}^2

nuts (%)

and cashew nuts. It must be taken into account that the fermented WCNEs did not present additives for their enrichment.

-0.35

0.773

NS

0.942

0.23

0.926

Sensory analysis of WCNE fermented with Lb. paracasei for ATCC 334

Table 2 presents the score averages of the sensory attributes by the acceptance test. F3 and F4, formulations with the highest cashew nut concentration, obtained the highest scores for all attributes, except for the appearance in F4. F3 differed significantly (P < 0.05) from the formulations with a lower concentration of cashew nuts (F1 and F2) in all attributes, except aroma. According to Tabanelli et al. (2018), volatile compounds such as aldehydes, alcohols, and aromatic amino acids are metabolites produced in the cashew nuts fermentation by lactic bacteria and provide an abundance of aroma.

F1 and F2 (Table 2) received the lowest scores in the flavor attribute (slightly disliked), evidencing a significant difference (P < 0.05) from other formulations, which reveals that lower proportions of cashew nuts directly affect the acceptance of WCNE. For the panelists, these formulations presented an acidic taste and low sugar concentration. Morais (2009) evaluated the acceptance of WCNE with different concentrations of sugar regarding appearance and aroma attributes. The authors reported that the degree of sweetness increases the acceptability of WCNE. Other studies with watersoluble plant extract also confirm this correlation (JAEKE et al, 2010). There were no significant differences (P < 0.05) between the formulations for the aroma attribute (Table 2).

Table 3 shows that the cashew nut concentration significantly affected (P < 0.05) all attributes, except for the global impression, which obtained satisfactory results. The response surfaces for the sensory analysis (Figure 2) confirmed that superior cashew nut concentrations lead to higher notes.

-0.17

0.681

NS

0.847

Global Impression

NS

0.87

NS

0.969

Figure 2 (A, B, and D) show that the attributes aroma, appearance, and global impression did not obtain significant influence (P < 0.05) from *Lb. paracasei*. Oppositely, the response for the flavor attribute reflects that lower probiotic concentrations are correlated to higher grades.

Acceptability by consumers is one of the biggest challenges for fermented plant-based products. The probiotics alter taste, aroma and texture aspects due to a diversity of metabolic components production, substrates, and fermentation periods (PANGHAL et al., 2017). According to Huang et al. (2017), adding flavorings to vegetable drinks may disguise the fermentation changes, achieving a better acceptance by the consumer.

For Kehinde et al. (2020), more studies with fermented water-soluble extracts and plant-based probiotic drinks are required since these products' organoleptic aspects must correlate to dairy beverage standards to present greater sensorial acceptability.

CONCLUSION

WCNE presented itself as a suitable fermentation substrate for *Lactobacillus paracasei* ATCC 334 with cell viability between 7.72 log CFU g⁻¹ to 8.27 log CFU g⁻¹, after 6 hours of fermentation. The concentration of cashew nuts (CNC) significantly influenced (P < 0.05) all physicochemical parameters



and sensory attributes of the fermented WCNE. The beverage has the potential to be a ready-todrink probiotic product under FDA regulations. It is important to emphasize that more studies should be carried out in the future to evaluate the shelf-life and toxicity of the drink for its availability on the market. Fermented WCNE can meet the probiotic demand of people on a milk-restricted diet.



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DECLARATION OF CONFLICT OF **INTEREST**

We have no conflict of interest to declare.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved the final version.

REFERENCES

AMORIM, M. et al. Use of whey peptide fraction in coated cashew nut as functional ingredient and salt replacer. LWT, v. 92, p. 204–211, 2018. Available from: https://doi.org/10.1016/j. lwt.2017.12.075>. Accessed: Jul. 06, 2019. doi: 10.1016/j. lwt.2017.12.075.

AOAC. Official Methods of Analysis, 20th ed. Washington D.C.: AOAC International. 2016.

APHA. Compendium of Methods for the Microbiological Examination of Foods. American Public Health Association. 4. ed. Washington: APHA, 2001.

BRASIL. Ministério da Saúde. Agência Nacional de Vigilância Sanitária. Resolução RDC nº 241, de 26 de julho de 2018. Critérios sobre os requisitos para comprovação da segurança e dos benefícios à saúde dos probióticos para uso

em alimentos. Diário Oficial da União, Poder Executivo, 27 de julho. 2018. Available from: http://portal.anvisa.gov.br/ documents/10181/3898888/RDC_241_2018_.pdf/941cda52-0657-46dd-af4b-47b4eF4335b7>. Accessed: Sep. 03, 2019.

BRASIL. Agência Nacional de Vigilância Sanitária. RDC nº 54, de12 de Novembro de 2012. Dispõe sobre o Regulamento TécnicosobreInformaçãoNutricionalComplementar.Availablefrom:<http://bvsms.saude.gov.br/bvs/saudelegis/anvisa/2012/</td>rdc0054_12_11_2012.html>.Accessed: Feb. 05, 2019.

BRAINER M. S. C.P.; VIDAL M. F. Cajucultura. Escritório técnico de Estudos Econimicos do nordeste – ETNENE. 2020. Available from: https://www.bnb.gov.br/ documents/80223/7106244/114_Caju.pdf/b0348238-45be-b060-3629-488c2e70a499>. Accessed: Jul. 14, 2020.

CODINA-TORRELLA, I. et al. Microbiological stabilization of tiger nuts' milk beverage using ultra-high pressure homogenization. A preliminary study on microbial shelf-life extension., Food Microbiology . 2018. Available from: https://doi.org/10.1016/j.fm.2017.08.002). Accessed: Jul. 06, 2019. doi: 10.1016/j. fm.2017.08.002.

FDA. Guidelines for industry. Early clinical trials with live biotherapeutic products: chemistry, manufacturing, and control information. 2003. Available from: https://www.fda.gov/downloads/Biologi.../UCM292704.pdf>. Accessed: Oct. 29, 2019.

GROM, L. C. et al. Probiotic dairy foods and postprandial glycemia: A mini-review, **Trends in Food Science; Technology**. 2020. Available from: https://doi.org/10.1016/j.tifs.2020.05.012 Accessed: Jul. 06, 2019. doi: 10.1016/j.tifs.2020.05.012.

GU, Y. et al. Impact of *Lactobacillus paracasei* IMC502 in coculture with traditional starters on volatile and non-volatile metabolite profiles in yogurt. **Process Biochemistry**. 2020. Available from: https://doi.org/10.1016/j.procbio.2020.07.003. Accessed: Oct. 23, 2020. doi: 10.1016/j.procbio.2020.07.003.

HUANG, S. et al. Spray drying of probiotics and other food grade bacteria: A review. **Trends in Food Science**; **Technology**, 63, 1–17. 2017. Available from: https://doi.org/10.1016/j.tifs.2017.02.007. Accessed: Oct. 23, 2020. doi: 10.1016/j.tifs.2017.02.007.

IAL-INSTITUTO ADOLFO LUTZ. Normas Analíticas do Instituto Adolfo Lutz: Métodos químicos e físicos para análise de alimentos. 4. ed. São Paulo: IMESP, 2008.

JAEKEL, L. Z. et al. Avaliação físico-química e sensorial de bebidas à base de extratos de soja e de arroz. **Ciênc.Tecnol.** Aliment., Campinas, 30(2): 342-346, abr.-jun. 2010.

KEHINDE, B. A. et al. Vegetable milk as probiotic and prebiotic foods. Advances in Food and Nutrition Research. 2020. Available from: https://doi.org/10.1016/bs.afmr.2020.06.003. Accessed: Oct. 23, 2020. doi: 10.1016/bs.afmr.2020.06.003.

MAPA. Ministério da Agricultura, Pecuária e Abastecimento. IN nº 19/2013. Estabelecer em todo território nacional a complementação dos padrões de identidade e qualidade para bebidas. Available from: https://alimentusconsultoria.com.br/ wp-content/uploads/2016/07/IN-19-2013.pdf>. Accessed: Jun. 09, 2020.

MOHAN, V. et al. Cashew Nut Consumption Increases HDL Cholesterol and Reduces Systolic Blood Pressure in Asian Indians with Type 2 Diabetes: A 12-Week Randomized Controlled Trial. **The Journal of Nutrition**, v.148, n.1, p.63–69, 2018. Available from: https://doi.org/10.1093/jn/nxx001. Accessed: Jun. 06, 2019. doi: 10.1093 / jn / nxx001.

MORAIS, A. C. S. et al. Seleção de julgadores e avaliação de diferença sensorial entre extratos hidrossolúveis da amêndoa da castanha de caju (*Anacardium occidentale* L.). **Boletim do Centro de Pesquisa e Processamento de Alimentos**, v.28, p.281-288, 2010. doi: 10.5380/cep.v28i2.20442.

NOROUZI, S. et al. A Survey on the survival of *Lactobacillus paracasei* in fermented and non-fermented frozen soy dessert. **Biocatálise e Biotecnologia Agrícola**, 101297. 2019. Available from: https://doi.org/10.1016/j.bcab.2019.101297. Accessed: Oct. 23, 2020. doi: 10.1016/j.bcab.2019.101297.

PANGHAL, A. et al. Potential non-dairy probiotic products – A healthy approach. **Food Bioscience**, v.21, p.80–89. 2017. Available from: https://doi.org/10.1016/j.fbio.2017.12.003. Accessed: Jun. 06, 2019. doi: 10.1016/j.fbio.2017.12.003.

REBOUÇAS, M. C. et al. Optimization of the acceptance of prebiotic beverage made from cashew nut kernels and passion fruit juice. **Journal of. Food Science**. v.79, n.7, p.1393-1398, 2014. Available from: https://doi.org/10.1111/1750-3841.12507. Accessed: Jun. 06, 2019. doi: 10.1111 / 1750-3841.12507.

SANTOS, M. C. M. et al. Fermentation of chickpea (*Cicer* arietinum L.) and coconut (*Coccus nucifera* L.) beverages by *Lactobacillus paracasei* sub sp paracasei LBC 81: The influence of sugar content on growth and stability during storage. **LWT**. 2020. Available from: https://doi.org/10.1016/j.lwt.2020.109834. Accessed: Oct. 23, 2020. doi: 10.1016/j.lwt.2020.109834.

SILVA, N. et al. Manual de métodos de análise microbiológica de alimentos e água. 5ª edição. Ed. Blucher. 2017.

TABANELLI, G. et al. Fermented Nut-Based Vegan Food: Characterization of a Homemade Product and Scale-Up to an Industrial Pilot-Scale Production. **Journal of Food Science**. 2018. Available from: https://doi.org/10.1111/1750-3841.14036. Accessed: Jun. 06, 2019. doi: 10.1111/1750-3841.14036.

ZIELIŃSKA, D.; KOLOŻYN-KRAJEWSKA, D. Food-origin lactic acid bacteria may exhibit probiotic properties. **BioMed Research International**, v.2018, 2018. Available from: https:// www.hindawi.com/journals/bmri/2018/5063185/>. Accessed: Sep. 18, 2021. doi: 10.1155/2018/5063185.

9