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Formulação e caracterização dos parâmetros de qualidade de geleias e doces em pastas de pitanga roxa (*Eugenia uniflora* L.) com redução de açúcares

Formulation and characterization of the quality parameters of jellies and candy pastes of purple pitanga (*Eugenia uniflora* L.) with reduced sugars

Formulación y caracterización de parámetros de calidad de jaleas y dulces en pastas de pitanga púrpura (*Eugenia uniflora* L.) con azúcares reducidos

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Kataryne Árabe Rimá de Oliveira
ORCID: https://orcid.org/0000-0002-4582-333X
Universidade Federal da Paraíba, Brasil
E-mail: katarynearabe_@hotmail.com
Ana Flavia Gomes de Medeiros
ORCID: https://orcid.org/0000-0001-6807-0070
Universidade Federal da Paraíba, Brasil
E-mail: anaflaviamedeiros3@gmail.com
Nathália Kelly Paulino Ferreira
ORCID: https://orcid.org/0000-0001-6940-6998
Universidade Federal da Paraíba, Brasil
E-mail: na.kelly@hotmail.com
Maria Helena Araújo de Vasconcelos
ORCID: https://orcid.org/0000-0002-7596-3528
Universidade Federal da Paraíba, Brasil
E-mail: helenanutricionista@hotmail.com
Laís Chantelle
ORCID: https://orcid.org/0000-0003-3295-2420
Universidade Federal da Paraíba, Brasil
E-mail: lais.chantele@hotmail.com
Isabelle de Lima Brito
ORCID: https://orcid.org/0000-0003-3295-2420
Universidade Federal da Paraíba, Brasil
E-mail: isa_limab@hotmail.com

Resumo

O objetivo do presente trabalho foi caracterizar e avaliar microbiologicamente diferentes formulações de geleias e doces em pasta de pitanga roxa com teores reduzidos de sacarose. As formulações elaboradas para geleias foram "geleia A" com a proporção de 100% e "geleia B" com proporção de 50%, todas adicionadas de pectina (1,5 g) e ácido cítrico (0,45 g). Para o doce em pasta as concentrações foram "doce A" com 60% e "doce B" com 40%, todas acrescidas de ácido cítrico (0,6 g) e água (36 mL). O processo de elaboração seguiu todas as normas de controle de qualidade e em sequência foram realizadas as caracterizações físico-químicas (umidade; teor de cinzas; sólidos solúveis; pH; acidez titulável; açúcares; parâmetros de cor; antocianinas; flavonóides) e microbiológicas (coliformes totais e termotolerantes; bolores e leveduras; *Estafilococus* coagulase positiva e *Salmonella* sp.). Os resultados das análises foram comparados apenas entre as formulações do mesmo tipo de produto. Todos os dados físico-químicos estavam dentro dos parâmetros normatizados para geleias e doces. Quanto ao aspecto cor, às amostras apresentaram um característico vermelho intenso, diferindo apenas nos parâmetros de cromaticidade e luminosidade em doces e geleias respectivamente. Os subprodutos da pitanga apresentaram ainda concentrações variadas de compostos bioativos, com destaque para maiores quantidades de antocianina. Em todos os aspectos microbiológicos avaliados, os resultados apresentaram conformidade com os padrões estabelecidos pela legislação. Assim, verifica-se a possibilidade do desenvolvimento de geleias e doces de pitanga roxa, com redução no teor de sacarose, mas com características nutricionais, microbiológicas e visuais interessantes.

Palavras-chave: *Eugenia uniflora* L.; Produtos de frutas; Propriedades funcionais; Parâmetros físico-químicos e microbiológicos.

Abstract

The objective of the present work was to characterize and microbiologically evaluate different formulations of jams and paste candies of purple pitanga with reduced sucrose contents. The formulations made for jams were "jam A" with a 100% proportion and "jam B" with a 50% proportion, all with the addition of pectin (1.5 g) and citric acid (0.45 g). For the paste candies the concentrations were "candy A" with 60% and "candy B" with 40%, all added with citric acid (0.6 g) and water (36 mL). The elaboration process followed all the quality control standards and, in sequence, the physicochemical (humidity; ash content; soluble solids; pH; titratable acidity; sugars; color parameters; anthocyanins; flavonoids) and microbiological (total and thermotolerant coliforms; molds and yeast; coagulase positive *Staphylococcus* and

Salmonella sp.) characterizations were performed. The results of the analyses have been compare only between formulations of the same type of product. All physicochemical data were within the standard parameters for candies and jams. As for the color aspect, the samples showed a characteristic intense red, differing only in the chromaticity and luminosity parameters in sweets and jellies respectively. The pitanga by-products presented varied concentrations of bioactive compounds, with emphasis on greater amounts of anthocyanin. For all microbiological aspects evaluated, the results were in accordance with the standards established by the legislation. Thus, the possibility of the development of purple pitanga jams and candies has been showned, with a reduction in sucrose content, but with interesting nutritional, microbiological and visual characteristics.

Keywords: *Eugenia uniflora* L.; Fruit products; Functional properties; Physicochemical and microbiological parameters.

Resumen

El objetivo del presente trabajo fue caracterizar y evaluar microbiológicamente diferentes formulaciones de jaleas y dulces en pasta de pitanga púrpura con niveles reducidos de sacarosa. Las formulaciones hechas para jaleas fueron "Jalea A" con una proporción del 100% y "Jalea B" con una proporción del 50%, todas añadidas con pectina (1,5 g) y ácido cítrico (0,45 g). Para la pasta dulce, las concentraciones fueron "dulce A" con 60% y "dulce B" con 40%, todas con ácido cítrico (0,6 g) y agua (36 ml). El proceso de elaboración siguió todas las normas de control de calidad y, en secuencia, las caracterizaciones físico-químicas (humedad; contenido de cenizas; sólidos solubles; pH; acidez titulable; azúcares; parámetros de color; antocianinas; flavonoides) y microbiológicos (coliformes totales y termotolerantes; mohos y levaduras; *Staphylococcus* coagulasa positivo y *Salmonella* sp. Los resultados de los análisis se compararon solo entre formulaciones del mismo tipo de producto. Todos los datos físico-químicos estaban dentro de los parámetros estandarizados para jaleas y dulces. En cuanto al aspecto del color, las muestras mostraron un rojo intenso característico, y solo diferían en los parámetros de cromaticidad y luminosidad en dulces y jaleas, respectivamente. Los subproductos de Pitanga también mostraron concentraciones variables de compuestos bioactivos, con énfasis en mayores cantidades de antocianina. En todos los aspectos microbiológicos evaluados, los resultados mostraron cumplimiento con los estándares establecidos por la legislación. Por lo tanto, existe la posibilidad del desarrollo de jaleas y dulces de pitanga púrpura, con contenido reducido de sacarosa, pero con interesantes características nutricionales, microbiológicas y visuales.

Palabras clave: *Eugenia uniflora* L.; Productos de frutas; Propiedades funcionales; Parámetros fisicoquímicos y microbiológicos.

1. Introduction

Native from Brazil, the pitangueira is a plant that adapts to the climatic and edaphic conditions of the Northeast region of Brazil, where it is largely produced. Its fruits have an exotic flavor, varied coloring, of high nutritional quality and bioactive substances, such as phenolic acids, flavonoids, anthocyanins, carotenoids and vitamins, responsible for its high antioxidant potential (Vinholes et al., 2018).

Consideration of the growing search of the population, for foods that aggregate functionality to the pleasant taste, the pitanga becomes an interesting fruit to be used for the formulation of by-products. This use is relevant, since the pitanga has a relatively short shelf life after harvest, needing techniques that allow its long-term use (Sanches et al., 2017). Therefore, pulps, candies, drinks, ice cream, jellies and liquors are examples of these by-products obtained aiming to minimize fruit losses, in addition to adding nutritional value and interesting sensory characteristics (Frazon et al., 2018).

Jellies and candies are products obtained by mixing ingredients in proportions and suitable conditions, prepared from the boiling of fruit pulp with added sugar, acid and gelling agent until to reach the desired content of soluble solids (Brasil, 1978). Both have great acceptance and commercial importance for the fruit conserves industry in Brazil (Sanches et al., 2017).

However, according to current legislation, candies must be made from a mixture of vegetable ingredients and sugars in proportion 1:1. The same ratio is determined for jellies type extras, the ones with the lowest sugar content in their formulation (Brasil, 1978). Nevertheless, sucrose consumption has been associated with the appearance of several diseases, being recommended its decreasing in the diet (Khan et al., 2019).

Given the above, the objective of the present work was to elaborate and to characterize the physicochemical and microbiological parameters of jellies and purple pitanga candies with reduced sucrose levels.

2. Materials and Methods

The study design of the present work is characterized as laboratory research of a quantitative nature, where part was carried out in the field, such as the collection of materials, and the other laboratory part, as the determination of physical-chemical, microbiological characteristics and statistical analysis (Pereira et al., 2018).

2.1. Raw materials

The pitangas of the purple variety had been acquire in different locations of rural and urban region of the municipality of Bananeiras, Paraíba, Brazil. The fruits had previously been collect, selected as to the stage of maturation (intermediate stage of development, healthy and spotless) and transported under suitable conditions (time and temperature) to the Product Development Laboratory of the Federal University of Paraíba.

Before rehearsals, the fruits had been washed with detergent and running water and the disinfected surface by immersion in sodium hypochlorite solution (150 ppm) for 15 min, washed with sterile distilled water and dried for 2 h in a biosafety cabinet.

After selection and cleaning the fruits were stored under freezing (-18 °C) until the completion of the stages of formulation of jellies and candies and analysis of quality parameters. Beyond fruits, demerara-type sucrose was used, citric acid (Modern chemistry, São Paulo, Brazil) and pectin with high methoxyl content (Danisco, São Paulo, Brazil).

2.2 Formulations of Jams and Paste Candies

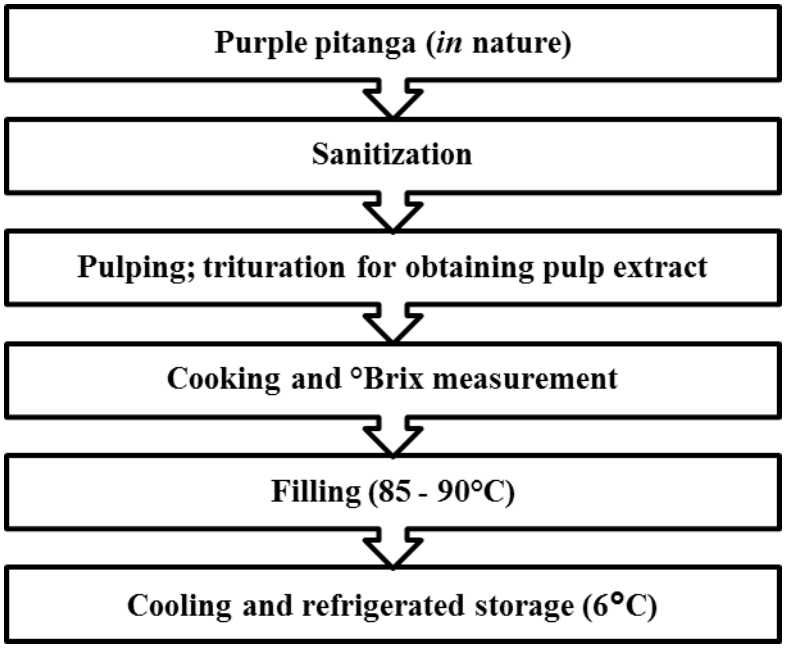
Previously sanitized pitangas were thawed for the pulping process of the fruit. The pulp obtained was crushed and the sieved input for the production of the final extract used in the formulation of jams and candies of purple pitangas.

Jellies and candies were obtained using different proportions of sugar (sucrose): “Jam A” with a 100% proportion (150 g of fruit pulp for 150 g of sugar) and “jam B” with a 50% proportion (150 g of fruit pulp for 75 g of sugar), all with the addition of commercial pectin (1.5 g) and citric acid (0.45 g); “Candy A” with 60% (100 g of fruit pulp for 60 g of sugar) and “candy B” with 40% (100 g of fruit pulp for 40 g of sugar), all added with citric acid (0.6 g) and water (36 mL).

The mixtures were subjected to heating, in open pan, with constant agitation, to prevent lumps from forming. Pectin was added to the jellies when boiling was reached (Curi et al., 2018).

After reaching 65 °Brix (jellies) and 60 °Brix (candy) (digital bench-top refractometer Milwaukee MA871 digital model), the products were packaged in sterile glass containers, with a capacity of 120 g, at a temperature of 85 - 90 ºC and then stored at 6 ° C until the moment of the physical-chemical and microbiological analyzes. The general flowchart of processing is also shown in Figure 1.

Figure 1. Processing flow chart for elaboration of jelly and candy pastes purple pitanga (*Eugenia uniflora* L.).



Source: Authors (2020).

As different products were processed, the description in the flowchart facilitates the visualization of all the steps, in sequence, necessary for production and final storage, making it extremely useful for reproducing the study.

2.3 Determination of physicochemical parameters

To characterize quality parameters in the jams and paste candies, the analyzes of total soluble solids (TSS), titratable acidity (TA), pH, total humidity, ash content, total sugars, reducers sugars, and color were performed.

TSS was determined using a digital refractometer Model Milwaukee MA871 (Milwaukee Instruments, Inc., Rocky Mount, NC, USA) and the results were expressed in °Brix (IAL, 2008). TA was determined by titration with NaOH (0,1 N) and use of phenolphthalein (1%) as indicator. Results of TA measurements were expressed in g of citric acid/100 g of product (IAL, 2008). pH values were measured with a digital pH meter (Model PHS-3E digital ion, Jiangshu Instruments, Jiangshu, China) (IAL, 2008).

For the analysis of humidity content, the desiccation method was used, by drying process in an oven at 105 ºC until constant weight, using 5g of the sample in previously tared and humidity-free porcelain crucibles (IAL, 2008). At the end of the humidity analysis, samples were used (3 to 5g), arranged in capsules, for ash determination, according to the incineration methodology (IAL, 2008). Total and reducing sugars were determined using the Eynon-Lane titration redox method (IAL, 2008).

The color was measured at room temperature using the electronic colorimeter GRETAG MACBETH, model COLOR-EYE 2180 (New Windsor, USA), with determination by the CIELab system: L* (0: dark, 100: white), a* (negative value: green, positive value: red) and b* (negative value: blue, positive value: yellow), in addition to the termination of C * (chroma) and tonality angle (anglo hue - h°) according to the International Lighting Commission (CIE, 1986).

2.4 Quantification of Anthocyanins and yellow flavonoids

Quantification of anthocyanins and yellow flavonoids was performed only on paste candies as an evaluative parameter for the products of derivatives of purple pitanga. An ethanol extracting solution was prepared - 1.5N HCl, in which one gram of the sample was dissolved in 50 mL of the extracting solution, remaining at rest, protected from light, under refrigeration, for a period of 16 hours. Subsequently the material, protected from light, was filtered using vacuum filtration read on a spectrophotometer in wavelength of 535 nm for anthocyanin and 374 nm for flavonoids. The results were calculated and expressed in mg of total anthocyanins/ 100mL (reading at 535 nm) and mg of flavonoids/100 mL (reading at 374 nm), according to equations 1 and 2, respectively (Francis, 1982).

Total anthocyanins = Dilution factor x absorbance / 98,2 Eq.1

Yellow flavonoids = Dilution factor x absorbance /76,6 Eq.2

2.5 Determination of microbiological parameters

Microbiological analyzes were performed to characterize the hygienic-sanitary parameters of the jellies and candies produced, following the standards of Brazilian legislation (Brazilian legislation, Technical Regulation on Microbiological Food Standards, 2001).

For count of total and thermotolerant coliforms, the Most Likely Number technique was used (MLN). Positive Coagulase *Staphylococcus* (PCS), *Salmonella* sp., molds, and yeasts were assessed using the classic standard plate counting method, determining the number of Colony Forming Units (CFU). Cultivation media and sowing techniques varied according to the investigated microorganism (Apha, 2015).

2.6 Statistical analysis

The analysis of the evaluation of the general quality parameters (physical-chemical and microbiological analysis) were performed in triplicate and all results were expressed as the average values of the data obtained.

Statistical analyzes were performed using descriptive statistics (mean and standard deviation) and inferential (ANOVA tests, followed by Student's tests) to determine differences ($p \leq 0,05$) between the results obtained. For statistical analysis, the Sigma Stat software. 3.5.

3. Results and Discussion

3.1 Physicochemical characterization

Jams and candies had a characteristic flavor and aroma of the purple pitanga. The cooking time required to obtain the final product was higher in formulations with reduced sugar, that is, with lower carbohydrate content for the gelation process.

Results of the characterization of the physical-chemical quality parameters of the jams and candies of purple pitanga are shown in Table 1.

Table 1. Characterization of physicochemical parameters of jelly and paste candies of purple pitanga (*Eugenia uniflora* L.) produced with different sugar concentrations.

Formulations	Humidity	Ash content (%)	pH	Soluble solids (°Brix)	Titratable acidity (g/100g)*	Total sugars (%)	Reducing sugars (%)
Jellies							
Jelly A (100%)	30,03 (±0,56)[b]	0,36 (±0,04)[a]	3,27 (±0,04)[a]	63,87 (±1,88)[a]	0,84 (±0,25)[a]	60,85 (±1,84)[a]	39,77 (±0,34)[a]
Jelly B (50%)	34,23 (±2,20)[a]	0,35 (±0,04)[a]	3,21 (±0,01)[a]	60,13 (±2,14)[a]	0,89 (±0,10)[a]	53,65 (±0,59)[b]	36,92 (±1,01)[b]
Paste candies							
Candy A (60%)	35,00 (±1,73)[a]	1,12 (±0,55)[a]	3,21 (±0,02)[a]	60,27 (±0,32)[a]	2,47 (±0,06)[a]	26,67 (±2,52)[b]	21,33 (±3,06)[a]
Candy B (40%)	36,33 (±1,16)[a]	0,99 (±0,21)[a]	3,13 (±0,02)[b]	59,47 (±2,74)[a]	2,63 (±0,23)[a]	21,33 (±4,93)[a]	17,67 (±1,53)[a]

*Titratable acidity (g/100g): (grams of citric acid per 100 grams of jam or candy).
[a-b]: Mean values (± standard deviation) in the same column with different subscript lowercase letters are significantly different ($p \leq 0.05$), based on Student test). Source: Authors (2020).

It is observed that the average of the values determined for the humidity of the jellies, even though different between formulations ($p \leq 0,05$), are within the range found for most fruit jellies. These values being also according to the quality standard established by the Brazilian legislation (Brazil, 1978), which indicates the maximum humidity content for fruit jellies lower to 38% (Barros et al., 2019). For paste candies this parameter did not vary between the different formulations.

It was found that the jellies in both formulations, presented average levels of soluble solids close to 65 ºBrix, contents that are usually found in conventional fruit jellies (62 °Brix) (Barros et al., 2019). However, due to reduced sugar content in candy formulations, soluble solids showed reduced values, ranging from 59 to 60 ° Brix.

The content of soluble solids is considered an indicator of the amount of sugars present in the food (Vieira et al., 2017). This relationship was observed for pitanga jellies in which the amounts of total sugars were relatively close to the soluble solids content. High levels of total soluble solids (TSS) associated with low water content and pH may favor the formation of crystallization of sucrose, which is responsible for improving the viscosity and texture of the product (Oliveira et al., 2019).

It has been found through the results that there was no significant difference between the two formulations of jams and candies in the parameter of fixed mineral residue (ash). However, both products showed higher values when compared to those found in fresh natural

fruit of 0.22% (Oliveira et al., 2006). This fact can be related to the composition of the plant matrix, as well as demerara sugar. According to Bettani et al. (2014), demerara sugar has higher levels of fixed mineral residue than refined sugars, and this may have contributed to the values obtained in this study.

Formulations presented pH classified as acids, with little difference between the candy samples (throughout processing). The acidic pH is characteristic of the pitanga fruit, as reported in previous studies (Oliveira et al., 2006). It is important to note that the optimal pH for gel formation varies between 3.0 and 3.4, depending on the final content of soluble solids. Therefore, citric acid has been used in the formulation of jellies to correct the initial pH the extracts, aiming adequate gelling in the final product (Vieira et al., 2017).

For acidity, candies and jellies showed higher levels when compared to those found by Curi et al. (2018) in jelly formulated with 100% pitanga (TA = 0.59). Acidity content has been attributed to the type of fruit, maturation stage and presence of organic acids. The acidity values tend to decrease when the maturation stage evolves.

It has been found that the levels of total sugars and reducers were higher in jelly and candy formulations A, which had higher concentrations of sucrose. This decrease for sugars (sucrose) added has a positive effect on the chemical and sensory quality of the jelly, as it avoids the extremely candy taste, emphasizing the characteristics of the fruit (Lago et al., 2006). For reducing sugar content, this difference can be explained due to the lower conversion of sucrose to glucose (inverted sugar) in products with reduced sugar (50%).

3.1.1 Color analysis

The color parameters of purple pitanga jellies and candies were evaluated through instrumental analysis using colorimeter, and the results are shown in Table 2. The luminosity values were similar for both samples tested.

Table 2. Color parameters of purple pitanga jellies and Candy (*Eugenia uniflora* L.) made with different concentrations of sugar.

Formulations	L	a*	b*	C*	H°
Jellies					
Jelly A (100%)	13,87 (±1,45)[a]	19,82(±1,89)[a]	9,49 (±1,06)[a]	22,00 (±1,68)[a]	25,69 (±0,10)[a]
Jelly B (50%)	16,64 (±1,09)[a]	9,08 (±1,76)[b]	3,96 (±0,83)[b]	9,91 (±1,94)[b]	23,54 (±0,33)[b]
Paste candies					
Candy A (60%)	24,31 (±0,25)[a]	8,33 (±0,31)[b]	2,54 (±0,30)[a]	8,71 (±0,36)[b]	16,95 (±1,46)[a]
Candy B (40%)	23,95 (±0,62)[a]	9,51 (±0,43)[a]	2,90 (±0,28)[a]	9,94 (±0,49)[a]	16,94 (±0,92)[a]

[a-b]: Mean values (± standard deviation) in the same column with different subscript lowercase letters are significantly different ($p \leq 0.05$), based on Student test). Source: Authors (2020).

The color parameter a (+) indicates that the prevalent color in the four samples was red. The color parameter b (+), indicating the presence of the yellow component, was higher in the formulation with greater amount of fruit pulp (100% jelly). This product also presented a less dark / red color, characterized by the values of C* (parameter related to color intensity) higher in this formulation (Aguiar et al., 2016). °Hue angle indicates the color tone, ranging from 0 ° (red), 90 ° (yellow), 180 ° (green) and 360 ° (blue). In this way, both samples remained close to the origin, indicating to be a red sample and corroborating with the parameter a* which indicated positive values inherent to the mentioned color.

3.1.2 Contents of anthocyanins and yellow flavonoids

The levels of yellow flavonoids and anthocyanins were determined in the candy formulations, described in Table 3.

Table 3. Anthocyanin and yellow flavonoid values present in the purple pitanga Candy (*Eugenia uniflora* L.) with reduced sugar content.

Formulations	Anthocyanin*	Yellow flavonoid **
Candy A (60%)	27,26 (±0,18)[b]	20,11 (±0,37)[a]
Candy B (40%)	29,15 (±0,25)[a]	18,58 (±0,30)[b]

[a-b] Mean values (± standard deviation) in the same column with different subscript lowercase letters are significantly different ($p \leq 0.05$), based on Student test).
* The results were calculated according to the methodology and expressed in (mg of total anthocyanins / 100mL) and **(mg of flavonoids / 100 mL). Source: Authors (2020).

According to the results found, the pitanga by-products presented varied concentrations of bioactive compounds. Among the pigments evaluated, anthocyanin was predominant in the samples tested, with values similar to those reported by Lima et al. (2002) who observed levels of total anthocyanins in the purple pitanga pulp of 26 mg.100 g^{-1}. This finding is characteristic of the fruit, since the major pigment of pitanga is anthocyanin, which increases its concentrations according to the maturation period (Lima et al., 2002).

The yellow flavonoid values were also similar to those found by Lima et al. (2002) in the purple pitanga pulp (18 mg.100 g^{-1}). According to Malaman et al. (2011), pitanga is rich in phenolic compounds and these are normally present in the form of esters, glycosides and many of them are flavonoids.

3.2 Microbiological analyses

Table 4 presents the results of the microbiological evaluation of the jellies and candies of the purple pitanga (*Eugenia uniflora* L.).

Table 4. Microbiological characteristics of purple pitanga jellies and Candy (*Eugenia uniflora* L.), made with different concentrations of sugar.

Formulations	Total coliforms	Thermotolerant coliforms	Molds and yeast	PCS	*Salmonella sp.*
Jellies					
Jelly A (100%)	<3,0 NMP/g	<3,0 NMP/g	< $1x10^2$ UFC/g	<$1x10^2$ UFC/g	Absent
Jelly Jam B (50%)	<3,0 NMP/g	<3,0 NMP/g	< $1x10^2$ UFC/g	<$1x10^2$ UFC/g	Absent
Paste candies					
Candy A (60%)	<3,0 NMP/g	<3,0 NMP/g	$2,5x10^2$	<$1x10^2$ UFC/g	Absent
Candy B (40%)	<3,0 NMP/g	<3,0 NMP/g	$4,3x10^2$	<$1x10^2$ UFC/g	Absent

* FCU: Colony Forming Units; NMP: Most Likely Number; MLN: *Positive Coagulase Staphylococcus (*PCS*)*. Source: Authors (2020).

All parameters analyzed were within the limits allowed by the DRC nº 12/ 2001 (BRASIL, 2001). The values found attended the stipulated by legislation for fruit products (jellies, candies, pulps): maximum of 104 UFC/g for molds and yeasts, maximum of 10 2 NMP/g for coliforms at 45 °C and absence of *Salmonella* sp in 25g of sample. In addition to

the absence of contamination by positive coagulase *Staphylococcus* and total coliforms (35°C).

According to Silva (2010), the absence of bacteria in certain foods may be due to conditions that disadvantage their survival, as low pH and strains injured by the manufacturing process, in this case, exposure of fruits to high temperatures. The high levels of soluble solids and the humidity obtained can also ensure the microbiological stability of the product (Vieira et al., 2017).

These data also suggest that there were adequate processing procedures, such as sanctification of the fruits and equipment used, in addition to the effectiveness of the conservation methods employed. The combination of all these characteristics allows the microbiological stability of the product, making it an alternative to prolong the useful life of fruits and their derivatives.

4. Conclusion and Suggestions

It was possible to verify that the fruit of the pitangueira has good potential for production jellies and candies, no need to add sugar in large proportions.

Thus, the jelly in proportion of 50% of sugar and the candy formulated with 40% sugar were characterized with optimal nutritional properties, due to decreased sugars (sucrose, glucose) and attractive color aspects. Furthermore, the processing performed was effective, obtaining products with interesting nutritional potential and physical-chemical and microbiological quality within normal standards.

However, it is necessary to continue the study in order to assess the sensory quality of jellies and paste candies, besides possible functional characteristics (e.g., antioxidant properties), derivatives to the biotive compounds present in the formulations.

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Percentage of contribution of each author in the manuscript

Kataryne Árabe Rimá de Oliveira – 20 %

Ana Flavia Gomes de Medeiros – 20 %

Nathália Kelly Paulino Ferreira – 20 %

Maria Helena Araújo de Vasconcelos – 10%

Laís Chantelle – 10%

Isabelle de Lima Brito – 20 %