



# SCIENTIFIC INITIATION

Stimulus for research and  
path to knowledge production

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Sérgio Luis Melo Uiroli  
Fernando Morais Rodrigues  
Paula Jucá de Sousa  
Paulo Uitoriano Dantas Pereira



Compartilhando conhecimento



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# **SCIENTIFIC INITIATION: STIMULUS FOR RESEARCH AND PATH TO KNOWLEDGE PRODUCTION**

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**SCIENTIFIC INITIATION:  
STIMULUS FOR RESEARCH AND PATH TO  
KNOWLEDGE PRODUCTION**

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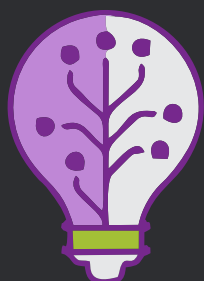
## **SCIENTIFIC INITIATION: STIMULUS FOR RESEARCH AND PATH TO KNOWLEDGE PRODUCTION**

### **Presentation**

The collection of scientific articles brings together articles authored by the teachers of the Undergraduate Courses in Chemistry, Mathematics and Food Technology at the Federal Institute of Education, Science and Technology of Tocantins IFTO – Paraíso do Tocantins campus. The scientific articles referring to the areas of exact and earth sciences, agrarian sciences and engineering, were prepared based on research and scientific initiation works developed, during the years 2018 and 2019, by teachers with the collaboration of scholarship students and volunteers, in order to contribute for academic quality, providing the ability to learn using the stimulus of scientific research and the production of knowledge.

The scientific works in this collection were presented at conferences, in an expanded summary format, allowing its authors to exchange experiences with researchers from Brazilian universities, members of the scientific communities in Brazil and abroad, on the dissemination and application of scientific research carried out. The publication of the collection reflects the commitment of the organizing professors with the technical-scientific bases and the development of scientific research in the undergraduate courses of the IFTO Campus Paraíso do Tocantins.

**MASTER SÉRGIO LUÍS MELO VIROLI**





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# SCIENTIFIC INITIATION

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# EXTRACTION AND EVALUATION OF MACAÚBA ALMOND OIL (*Acrocomia aculeata*) COLLECTED IN THE PARKING OF THE FEDERAL INSTITUTE OF TOCANTINS - IFTO CAMPUS PARAÍSO DO TOCANTINS

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## ABSTRACT

*Vegetable oils are liquid substances insoluble in water (hydrophobic), produced by many oil plants and used by various sectors, such as in the chemical, pharmaceutical, cosmetic and food industries. The objective of this work was the extraction and biometric characterization of the macauba. Samples of macauba coconut collected were in the parking lot of the IFTO campus Paraíso do Tocantins in the city of Paraíso do Tocantins. The biometric characterization of macauba carried was out using the methodology proposed by Manfio. The oil extracted was by a manual press and subjected to physico-chemical analyses of*

*moisture, acidity index, hydrogenionic potential-pH, peroxide index, saponification index and moisture following methodologies described by the Adolfo Lutz Institute. The results found indicated the feasibility of exploring macauba as an energy species and the possibility of obtaining oils with food quality and for the production of biodiesel.*

### **Keywords:**

*Biometrics; Oilseed; Manual Pressing.*



## INTRODUCTION

**V**egetable oils are liquid substances insoluble in water (hydrophobic), produced by many oil plants and used by various sectors, such as in the chemical, pharmaceutical, cosmetic and food industries (GUEDES, 2006; SILVA, 2009).

Macauba (*Acrocomia aculeata*) is a palm tree (Figure 01) native to tropical forests, which has a wide spread with natural stands in almost all Brazilian territory, but with extensive concentrations located in Minas Gerais, Goiás, Mato Grosso, Mato Grosso do Sul and the Brazilian Cerrado (LORENZI, 2006; CICONINI, 2012).



Figure 01. Macauba palm.  
Source: Portal da macauba, 2017.

*Acrocomia aculeata* is popularly known as macauba, macaíba, macaiúva, mocajá, mocujá, mucajá, bacaiúva, bocaiuva, coco-de-phlegm or coco-de-espinho, imbocaiá, umbocaiuva, depending on the region of its occurrence (LORENZI, MATOS, 2002). Its fruits are spherical with a diameter varying from 2.5 to 5.0 cm, 20% of skin (epicarp), 40% of pulp (mesocarp), 33% of chestnut (endocarp) and 7% of almond. The epicarp has a yellowish-brown color and breaks easily when ripe. The mesocarp is edible, fibrous, with a sweet taste, yellow or whitish in color. The endocarp is strongly adhered to the pulp, being very rigid and black in color. The almond is an edible oil that remains surrounded by the endocarp (CICONINI, 2012). Figure 02 illustrates the physical conformation of the macauba coconut.



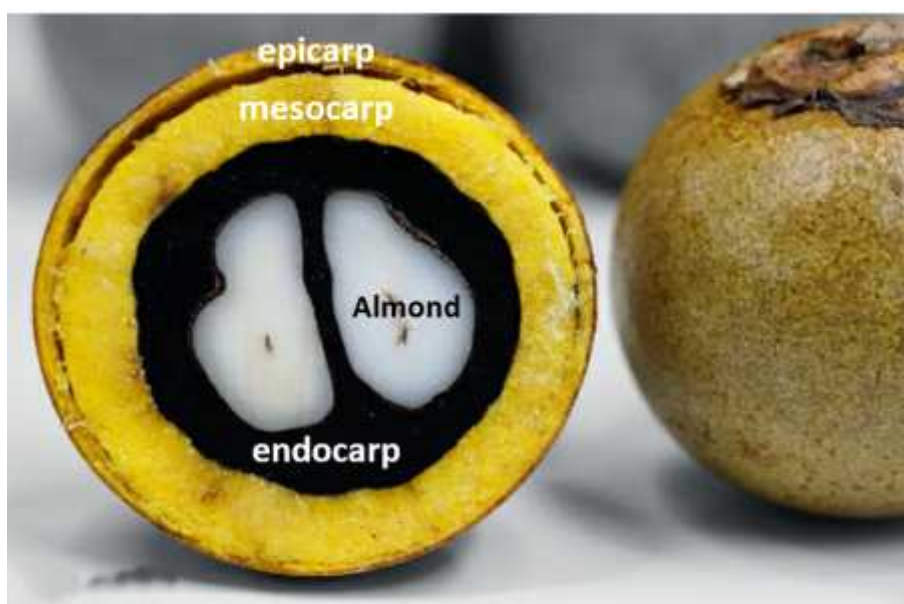


Figure 02. Parts of the macauba coconut.  
Source: Adaptation of the macauba portal, 2020.

Macauba has an oil productivity in the fruit in the range of 50-60% on a dry basis and 20-25% on a wet basis, with a productivity potential of up to 6000 kg of oil per hectare. Thus, the macauba palm can be a source of vegetable oils for the sectors of energy production, food and cosmetics (KALTNER et al., 2004; SILVA, 2007; MELO, 2012). Different extraction processes, including mechanical extraction, can obtain Macauba oil. Mechanical pressing is the most popular method for removing oil from oilseeds.

This extraction method offers greater security, simplicity of the process, favors the quality of crude oil and there is no presence of chemical residues, both for the oil and for the cake (PIMENTA, 2010; CICONINI, 2012; MELO, 2012). The oil extracted from the almond has a high content of lauric acid (38-45%) reaching high values in the market, used being in the cosmetics industry and later for the production of biokerosene, targeting the aviation sector (LIMA et al., 2007; SILVA, 2009). The objective of this work was the biometric characterization of the macauba, extraction by manual press and characterization of the oil of the macauba almond originating from the agroindustrial district of the City of Paraíso do Tocantins.

## METHODOLOGY

The experiment was carried out at the Federal Institute of Education, Science and Technology of Tocantins (IFTO) - Paraíso do Tocantins *Campus*. The extraction of the oil was carried out at the fruit and vegetable processing unit and the physico-chemical analyzes at the Analytical Chemistry Laboratory of the IFTO.



### Sample collection location

In January and February 2019, 11 kilos of the macauba fruit were collected in the IFTO parking lot (figure 03), located in the agro-industrial district of the municipality of Paraíso do Tocantins.



Figure 03. IFTO campus Paraíso do Tocantins parking

Source: Author

### Biometric Analysis

The biometric characterization of macauba was carried out using the methodology proposed by Manfio et al. (2011) with twenty fruits chosen at random, where the following characteristics were measured: fruit mass (g), almond mass (g) on an electronic scale with 0.01g precision, longitudinal diameter (cm), transversal of the fruit (cm) with precision caliper and fruit volume in a beaker (mL).

### Almond oil extraction route

The fruits transported were to the fruit and vegetable processing unit, where they underwent a selection process regarding the state of ripeness and integrity with the elimination of those that were apparently damaged or the appearance of injuries. After the evaluation, approximately 10 kilos of fruits selected were for oil extraction. The selected fruits previously cleaned were to remove dirt with the use of water and detergent.

After the cleaning process, they were subjected to a hygienization with sodium hypochlorite solution with a concentration of 100 milligrams per liter. After 30 minutes, the fruits were removed from the





sanitizing solution, subjected to a drying process and stored in a cool, ventilated place to avoid rotting of the fruits. The almonds were separated manually, crushed and heated to 105 ° C for 1 hour. Figures 04 and 05 illustrate the oil extraction route and the idealized press used for oil extraction, respectively.

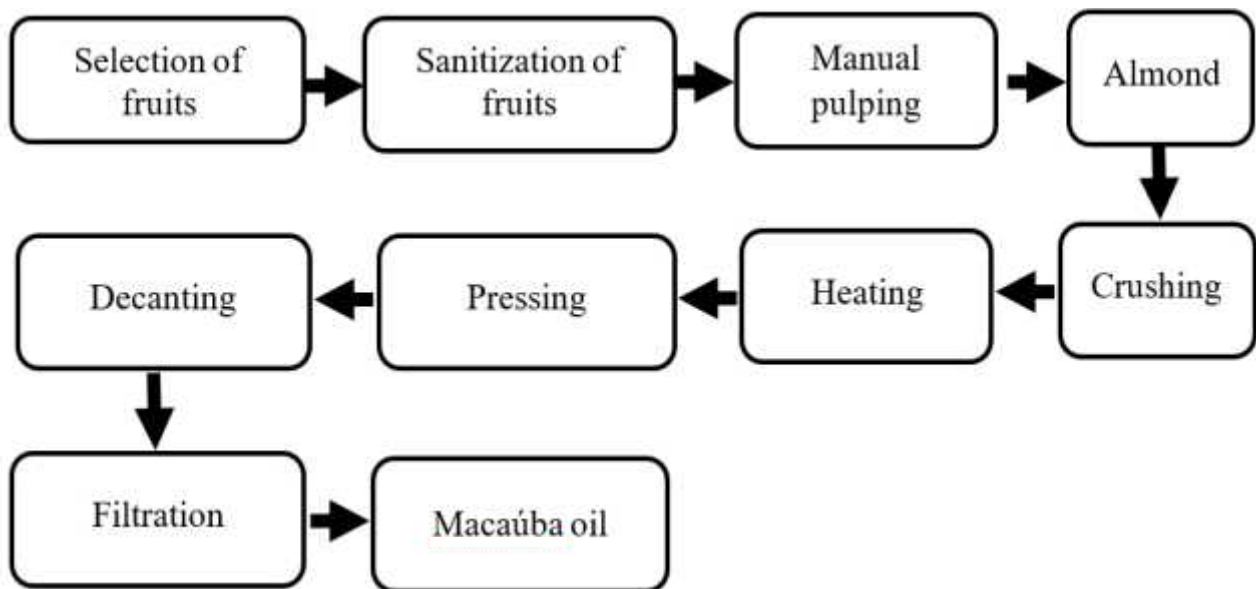


Figure 04. Flowchart of the route to obtain macauba oil.  
Source: Adaptation Silva and Caño Andrade (2011).



Figure 05. Press Manual designed for macauba oil extracting.  
Source: Author



## Physico-Chemical Analysis

The extracted oil was subjected to physical-chemical analysis of acidity index, hydrogenionic potential - pH, peroxide index, saponification index and moisture following the methodologies described by the Adolfo Lutz Institute (2004).

### Acidity index

The acidity was determined by weighing 2g of the sample in a 125 ml Erlenmeyer flask, containing 25 ml of ether and ethanol solution in a proportion of 2:1 respectively, followed by titration with standard 0.1 mol sodium hydroxide solution / L or 0.01 mol / L, in the presence of 1% phenolphthalein alcoholic solution as an indicator of titration.

### Hydrogenionic potential- pH

The reading of the pH measurement was carried out directly with the introduction of the bench digital pH meter electrode in 50 ml of the oil extracted from the macauba almond in a 100 ml Becher glass.

### Peroxide index

The determination of the peroxide index consisted of weighing  $5 \pm 0.05$  g of the sample in a 125 ml flask. 30 ml of 3:1 acetic acid and chloroform solution was added and stirred until completely dissolved. 0.5 ml of saturated KI solution was added and left to stand in the dark for one minute. Added 30 mL of distilled water and titrated with 0.01 mol/L sodium thiosulfate solution, with constant stirring, until the yellow color almost disappeared. Added 0.5 ml of starch indicator solution and continued titration until the blue color had completely disappeared. In this analysis, it was necessary to prepare a blank test under the same analysis conditions.

### Saponification index

The method for determining the saponification index was carried out using 5g of the sample in a flattened Erlenmeyer flask, with the addition of 50 mL of 4% alcoholic potassium hydroxide solution. A blank test was prepared and the analysis was carried out simultaneously with the sample. A condenser was connected and allowed to boil gently until the sample was completely saponified. Then, the system was cooled and the condenser was washed with a few milliliters of distilled water. The conical flask was disconnected from the condenser and the sample was titrated with a standard 0.5 mol/L hydrochloric acid solution, using two drops of 1% phenolphthalein alcoholic solution as an indicator of the titration.



## Moisture

Moisture was carried out by weighing  $\pm 5$  grams of sample added in a porcelain capsule, previously tared after calcination in a muffle at  $600^\circ\text{C}$ . Then they were placed in a drying oven at  $105^\circ\text{C}$  for 3 hours. Then they were removed and packed in a desiccator until reaching room temperature and subjected to weighing.

## RESULTS AND DISCUSSION

The results of the biometric analysis of the macauba fruit are in table 1.

biometric characteristic	Average
fruit mass (g)	$28.97 \pm 2.20$
almond mass (g)	$2.19 \pm 0.16$
Longitudinal fruit diameter (cm)	$3.65 \pm 0.16$
transverse fruit diameter (cm)	$4.18 \pm 0.17$
Volume of fruit (mL)	$23.50 \pm 4.57$

Table 01. Biometric characteristics of macauba fruits

The evaluation of these parameters helps in the layout of a production process, aiming to find a standard, which makes it possible to design equipment for industry, facilitating the process of separating the fruit components. The value found for the average mass of macauba  $28.97 \pm 2.20$  g is close to that described in the literature by Manfio et al. (2011), which obtained an average weight of  $32.1 \pm 16.53$  g and higher than those found by Machado (2015) for the average mass of  $19.67 \pm 3.19$  g. The analyzed macauba have a longitudinal external diameter equal to  $3.65 \pm 0.16$  cm and a transversal external diameter equal to  $4.18 \pm 0.17$  cm, values slightly higher than those found by Ciconini et al. (2012), 3.61 cm and 3.86 cm, and by Sanjinez-Argadonã (2011), 3.31 cm and 3.16 cm. The volume found is lower than those described by Manfio et al (2011). In which values of 36.68 ml found were, being between both values of the authors. Table 02 shows the results of the physico-chemical analysis of the oil extracted from the almond.



Physical-chemical analysis	Average	RDC N° 270/2005
Acidity index (mg KOH g <sup>-1</sup> )	0.69 ± 0.20	4.0
Hydrogenionic Potential (pH)	5.75	
Moisture (%)	16 ± 0.10	-----
Peroxide index (meq Kg <sup>-1</sup> )	5.12 ± 0.50	15
Saponification index (mg KOH <sup>-1</sup> )	191.58 ± 2.01	-----

Table 02. Physico-chemical analysis of the extracted oil.

The acidity index for the almond oil analyzed in this study was  $0.69 \pm 0.20$  mg KOH/g oil. Brazil (2005) establishes that cold-pressed oils, for human consumption, and not refined, must have a maximum of 4.0 mg KOH/g. Thus, the oil of the macauba almond would be within the regulation. Fernandes (2010) specifies that for biodiesel production, oils with moisture content below 0.5% and acidity below 2 mg KOH/g oil should be used. The oil extracted and analyzed in this study meets the specifications to use in the trans esterification process for the production of biodiesel.

The oil extracted from the macauba almond showed a pH equal to 5.71. The pH is a characteristic that the cosmetics and beauty market values and according to Fasina et al. (2006), the oil extracted from the macauba almond play a functional and sensory role in cosmetics, transport of fat-soluble vitamins (E, A, K and D) and supply of linoleic and linoleic acids.

Souza (2014), evaluating the evolution of the moisture of the macauba almond of the fruits collected between June and February found variation between 15.5% to 82.0% at different stages of maturation. Costa (2016) also evaluating the moisture content in macauba almonds found an average value of 10.14%. The value found in this research differs from Costa (2016) and Souza (2014) due to the stage of maturation of the macuba almond used for moisture analysis because as the fruit ripens, it tends to decrease the water content. Low moisture almonds and high oil content favor mechanical extraction, enabling the use of presses. The oil extracted from the almond showed an average moisture of 16%. According to Amaral (2007), the production of biodiesel can be negatively influenced by the presence of moisture in the oils in the trans esterification process with the deactivation of basic catalysts and the release of water molecules, decreasing the yield.

The peroxide index found in the oil extracted from the macauba almond in this study was  $5.12 \pm 0.50$  meq/Kg. According to Brazil (2005), the peroxide index established for cold-pressed and unrefined oils must be less than 15 meq/Kg. Studies by Hiane et al. (2005) did not detect peroxides in the analyzed samples, while Rodrigues (2007) found 8.0 meq/Kg for the peroxide index. AMARAL (2007) reached a value of 15.28 meq/Kg for the peroxide index in oil extracted from the macauba



almond. However, care should be taken to consider that the absence or low level of peroxide does not necessarily indicate that the oil has not undergone oxidation reactions, since the determination may occur in the period of decomposition of the peroxides, which are unstable and may be turned into by-products.

The saponification index varies inversely with the nature of the constituent fatty acids, that is, the lower the molecular weight, the greater the saponification index (MORETTO; FETT, 1998). In this study the saponification index value was 191.58mg KOH/g, being lower than the values of 221 mg KOH<sup>-1</sup>, 259 mg KOH/g and 308mg KOH/g, found by CETEC (1983), Hiane et al. (2005) and Amaral (2007), respectively. Considering the index of saponification of refining oils between 181 and 265mg KOH/g, the result of 191.58mg KOH/g then complies with the saponification index (HIANE et al., 2005).

## CONCLUSION

The biometric characterization of macauba indicated satisfactory values in relation to the fruits of macauba trees from other regions; these biometric characteristics enhance the extraction of its oils. The extracted oil presented values for the analysis of acidity and peroxide index within the limits permitted by Resolution No. 270/2005 of the National Health Surveillance Agency. The study indicates the feasibility of exploring macauba as an energy species and the possibility of obtaining oils with food quality and for the production of biodiesel.



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# CHARACTERIZATION OF THE BIODIESEL PRODUCED WITH OIL EXTRACTED FROM THE MACAUBA ALMOND BY METHYL TRANSESTERIFICATION WITH BASIC CATALYSIS

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## ABSTRACT

*Brazilian oilseed production stands out, in relation to other countries, due to its biodiversity and great availability of cultivation area. Macauba (*Acrocomia aculeata*, when pressed, oil is extracted in liquid phase at room temperature present in the pulp or the almond. The oils of the almond and the pulp of the macauba (*Acrocomia aculeata*) are promising alternatives in the production of biodiesel due to its high energy efficiency. With the current search for alternative sources of energy, macauba is one of the main native species with high potential for supplying oil for the production of biofuel. The objective of this work was to characterize the biodiesel produced with oil extracted from the macauba almond (*Acrocomia aculeata*) purchased at the local trade, covered fair in the City of Paraíso do Tocantins, produced by hand by small regional producers. The production of biofuel started with physical*

*chemical analysis, heating and transesterification of the oil. The biofuel obtained was analyzed according to the physical chemical parameters of visual aspect, dice acidity, specific mass, moisture and flame test following the recommendations of the physical-chemical analysis of the National Agency of Petroleum, Natural Gas and Biofuels - ANP through ANP Resolution No. 45 OF 08/25/2014. The biofuel produced met the requirements of the ANP, demonstrating the viability of production. Through the physical-chemical parameters analyzed, it was possible to verify that the biofuel produced presented satisfactory quality.*

### **Keywords:**

*Extractivism; Renewable Energy; Environmental Impact.*



## INTRODUCTION



The species *Acrocomia aculeata*, popularly known as macauba, is a palm tree native to tropical forests with wide distribution in the Brazilian territory, forming part of the rural landscape of several states and of the Cerrado and Pantanal Brazilian biomes (LORENZI, NEGRELLE, 2006; MOREIRA; SOUSA, 2009, RIBEIRO et al. 2017).

Macauba is a raw material used for the extraction of vegetable oil and the production of biofuel. Its fruit has approximately 35% moisture, weighing an average of 18 g when dry and is composed of four distinct parts: 19.77% of external skin (epicarp), 41.17% of oily mass (mesocarp), 28.97% woody bark (endocarp) and 10.09% oily almonds. Figure 01 illustrates the parts of the macauba

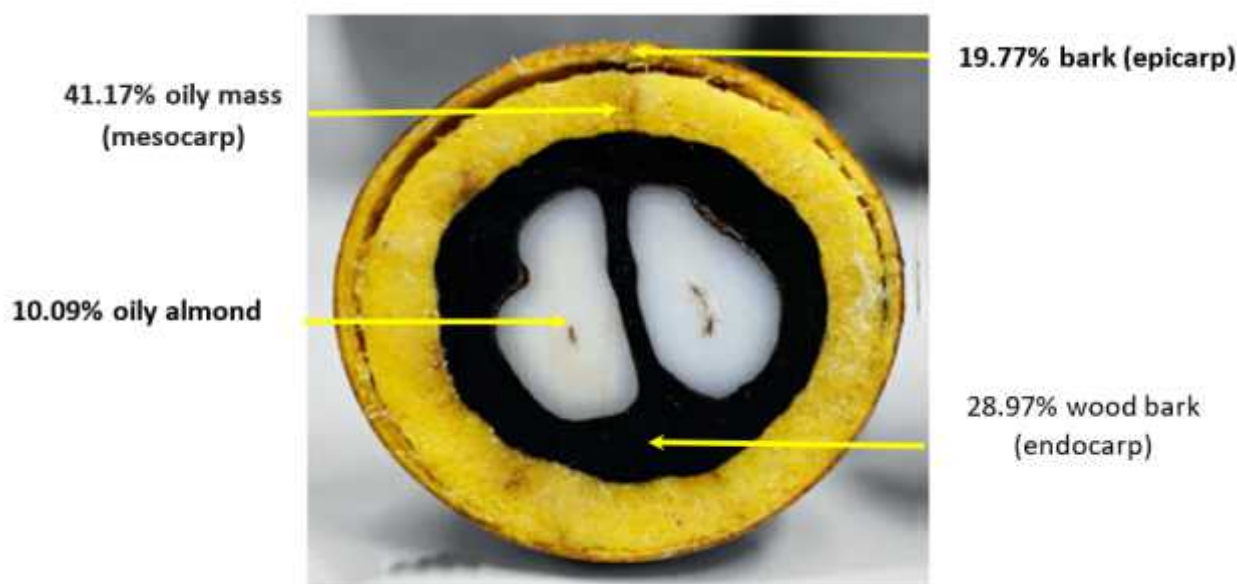


Figure 01. Parts of the macauba coconut.  
Source: adaptation of the macauba portal, 2020

The oil extracted from the macauba, concentrated in the mesocarp and almond, is used for the production of biodiesel, due to its great stability to oxidation and operability at low temperatures (CARVALHO, 2015). From this fruit, all parts can be used: the peel and the endocarp as fuel for burning biomass (BHERING, 2019). With the current search for alternative sources of energy, macauba is one of the main native species with high potential for supplying oil for the production of biofuel, as it produces up to 4,000 liters of oil per hectare per year, approaching yield palm oil, while soy, which is an annual crop, produces 420 liters per hectare per year (NETHERLANDS, 2004; TELES, 2009; CESAR et al. 2015). Furthermore, the oil produced by Macauba has no tradition of being a food product and has several industrial and energy applications (MICHELIN et al., 2015).



The use of macauba for biofuel production generates by-products that are highly valued for animal nutrition (COLLARES, FERREIRA; CABRAL, 2009; BOAS et.al., 2010). Macauba oil can be obtained by different extraction processes, including mechanical extraction. Mechanical pressing is the most popular method for removing oil from oilseeds. This extraction method offers greater security, simplicity of the process, favors the quality of crude oil and there is no presence of chemical residues, both for the oil and for the cake (PIMENTA, 2010; CICONINI, 2012; MELO, 2012).

The study of biofuel production from macauba is interesting since the cost of pulp oil is low, its extraction is easy and its cultivation, like other plants used for oil production, acts as a support to family farming creating better conditions of life in needy regions and offering alternatives to economic and socio-environmental problems (RAMOS, 2003).

We chemically define biodiesel as an ester of long-chain fatty acid, derived from renewable lipid sources, such as vegetable oils or animal fats. Biodiesel is a non-toxic, renewable and non-fossil alternative fuel. It consists of alkyl esters and has excellent lubricating characteristics, being able to supply all or part of the diesel oil derived from petroleum in diesel cycle engines, with minimal or no adaptation (KNOTHE, 2010; SANTACESARIA et al., 2012). It can be produced from the processing of oils and fats by transesterification reactions or by the esterification of fatty materials.

Transesterification or alcoholysis is a reaction process that consists of the reaction of triacylglycerols with a short carbon chain alcohol, in the presence of a homogeneous catalyst, such as sodium hydroxide (NaOH) or potassium hydroxide (KOH), forming alkyl monoesters and as a by-product glycerin or glycerol (LAM; LEE; MOHAMED, 2010). Figure 02 illustrates a transesterification reaction.

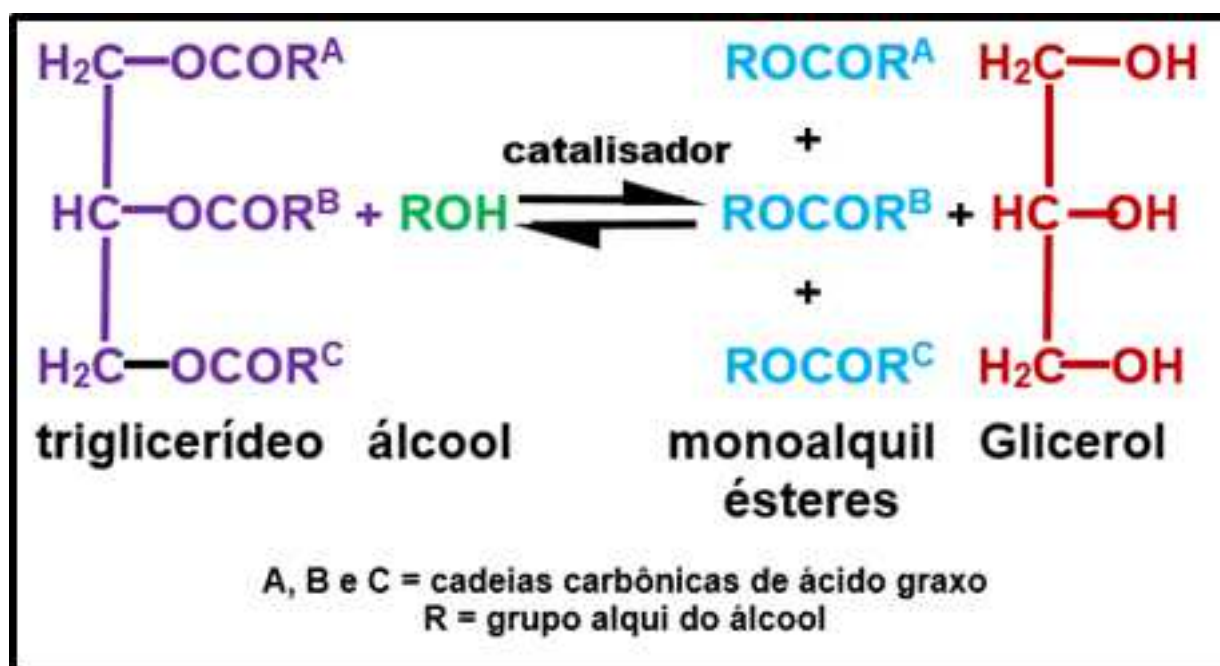


Figure 02. Transesterification reaction

Source: Author



Torically the transesterification must be completed with the biofuel showing high purity, no fatty acids, residual catalyst or excess alcohol from the reaction and traces of glycerol. The reaction occurs stoichiometrically with three moles of alcohol for each mole of triglyceride. In practice, the trans-spherical process generates biodiesel and glycerol, since excess alcohol used is to shift the reaction balance, maximizing the biodiesel yield and separating the glycerol formed during the rational process (KNOTHE et al, 2006).

The industrial process chosen for obtaining biodiesel is homogeneous alkaline transesterification using sodium hydroxide or potassium hydroxide and methanol and ethanol alcohols for their low cost, short carbon chains and great availability (FELIZARDO; CORREIA; RAPOSO, 2006)

The objective of this work is to characterize the biodiesel produced with the oil extracted from the macauba almond sold in the covered fair in the Municipality of Paraiso do Tocantins.

## METHODOLOGY

### Characterization of the oil extracted from the macauba almond

At the local market in the city of Paraiso do Tocantins, a covered fair, 500 mL of macauba almond oil extracted from artisanal production by small farmers in the region of the Middle Araguaia Valley were purchased at the local market. The oil sample taken was to the Analytical Chemistry laboratory of the Tocantins Science and Technology Education Institute - IFTO Paraiso of Tocantins campus for physical and chemical analysis. The oil was subjected to filtration processes to remove dirt and foreign matter. In addition, heating at 80 ° C and homogenization for 4 hours to remove possible amounts of water that may have accumulated in the oil (figure 03).



Figure 03. Oil extracting from the macauba almond under heating

Source: author





The methodology used to determine the acidity index, relative density at 25°C, peroxide index, saponification index and moisture content were carried out in triplicate following the determinations of the analytical standards of the Adolfo Lutz Institute (IAL, 2008).

### Production and characterization of biodiesel

The oil extracted from the macauba almond acquired in the local market was subjected to the process of heating at 80 ° C and homogenization for 5 hours to remove possible amounts of water that may have accumulated in the oil. The biodiesel produced with oil extracted from the macauba almond was analyzed according to the physical chemical parameters, Index Acidity mg KOH/g, Specific mass kg.m<sup>-3</sup> and Moisture content mg/kg, followed the methodology proposed by the Adolfo Lutz Institute (IAL, 2008). The method used to determine the flame test was carried out according to the methodology suggested by Geris (2007) and Santos Junior et. al. (2015).

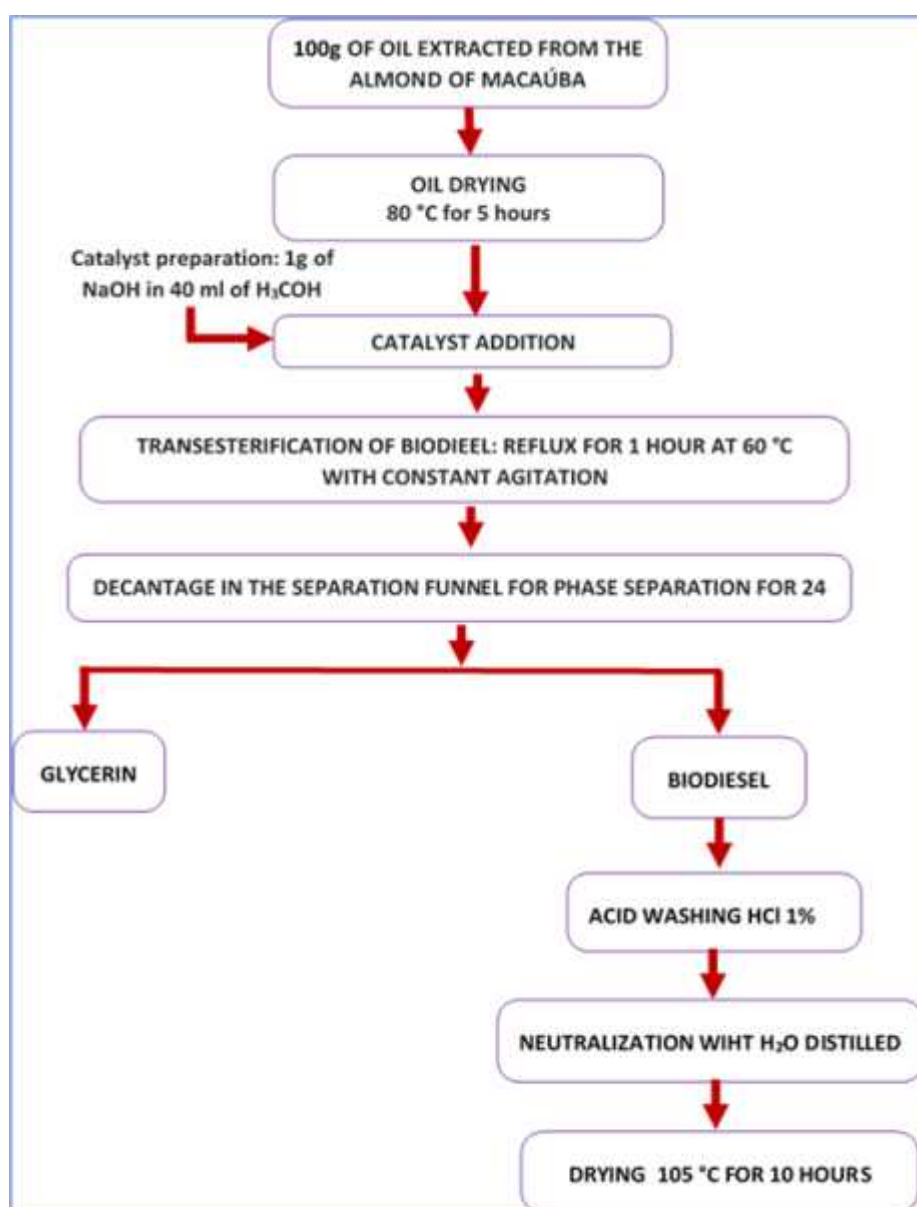


Figure 04. Flowchart of biodiesel preparation  
Source: Author



The visual aspect realized was through the observation of the biodiesel produced. The Experiment was conducted in three repetitions with only the mean between repetitions being analyzed. The results were compared with the limits required by the specification for biofuel established by ANP Resolution No. 45 of 08/25/2014 National Agency of Petroleum, Natural Gas and Biofuels published in the Official Gazette of the Union 26 August 2014 (BRASIL, 2014). The production process of biofuel followed the methodology proposed by Santiago (2018) and the steps described by the flowchart shown in Figure 04.

The results of the physical chemical analysis of the oil and biodiesel were analyzed through ANOVA analysis of variance and Tukey's test adopting the level of 5% probability ( $p \leq 0.05$ ), which was carried out with the Sisvar version 5.6 program (FERREIRA, 2011).

## RESULTS AND DISCUSSION

The results of the analysis of the oil extracted from the macauba almond are described in table 01.

Parameters Physicochemical	Oil extracted from macauba almond	IN N° 87, of march 15, 2021	ANP N°. 45 of august 25, 2014
Especific mass a 20°C (g/mL)	898.51 ± 6,70	-----	850 a 900
Acidity index (mgKOH/g)	0.48 ± 0.04	4.00	0.50
Peroxide index (meq.kg <sup>-1</sup> )	6.83 ± 2.02	15.00	-----
Saponification index (gKOH/100g)	206.15 ± 24.53	-----	-
Moisture mg/kg	186 ± 6.52	-----	-- ≤ 200.0

Table 01: Physico-chemical parameters of macauba almond oil.

According to the norms stipulated by the National Health Surveillance Agency ANVISA Normative instruction - IN No. 87, of March 15, 2021 published in the Official Gazette of the Federal Government DOU No. 51, of March 17, 2021 (BRAZIL, 2021) and Agency National Petroleum, Natural Gas and Biofuels - ANP through ANP Resolution No. 45 of August 25, 2014 (BRAZIL, 2014) the physical-chemical parameters analyzed shown in table 01, are in accordance with the respective legislation. Viroli et al. (2019), carrying out studies on the characterization of macauba almond oil extracted by mechanical pressing and Hiane et al., (2005), studying Pulp oil and macauba almond found results for the physical and chemical parameters analyzed in accordance with legislation, which demonstrates the relevance of knowledge of the physical and chemical characteristics of the raw material and the use of efficient processing.

Knowledge of the physical and chemical characteristics of vegetable and animal oils and fats used as raw material is essential to define the routes for efficient transesterification of biodiesel. Raw





materials with adequate chemical composition and within the specified standards are required to meet the quality parameters of biodiesel in physical-chemical terms. The parameters defined are technically and scientifically, allowing the prediction of the use of the raw material for the production of a biodiesel that meets the desired quality parameters, based on its chemical composition.

Biodiesel outdoes after the entire production and purification process showed physical and chemical results described in table 02.

Parameters Physicochemical	Biodiesel produced with macauba almond oil	ANP N°. 45 of august 25, 2014
Aspect	Clear and free of impurities	-----
Acidity index (mgKOH/g)	$0.46 \pm 0,03$	0.50
Especific mass a 20°C (g/mL)	$897.80 \pm 17.53$	850 a 900
Moisture mg/kg	$177.14 \pm 14.66$	$\leq 200.0$
Flame test	yellowish	----- ---

Table 02: Physical and chemical parameters of biodiesel produced with macauba almond oil.

According to ANP Resolution No. 45 of August 25, 2014 (BRASIL, 2014), biofuel presented physico-chemical parameters in accordance with the respective legislation. High levels of these parameters can make biofuel production unviable, so it is preferable to use oils with low levels of acidity (DOMINGOS, 2009). The flame test, based on the burning of the fuel obtained and the characteristic color of the yellow flame, showed a positive result confirming the flammability of the biofuel produced. The biofuel produced showed a clear yellow brown color. Carvalho et al (2014) analyzing the physical-chemical properties of biodiesel produced from macauba mesocarp oil via basic catalysis (*Acrocomia aculeata* Jacq.)

Obtained an average of 0.71 mg KOH g<sup>-1</sup> for the acidity index and 898.50 Kg m<sup>3</sup> for the specific mass. According to the parameters analyzed by the authors, the acidity index is outside the limit required by the specification for the biofuel established by ANP Resolution N ° 14 which determines the maximum limit of 0.5 mg KOH g<sup>-1</sup> of the sample.

The high acidity index makes the transesterification reaction difficult, causing corrosion in the engine and deterioration of the biofuel. A possible solution to reduce the index would be to subject the biofuel to another stage of transesterification via acid catalysis, in order to esterify the fatty acids still present in the medium (CANDEIA, 2008; MELO, 2012). The knowledge of the specific mass of a fuel allows us to evaluate the operation of the injection pumps in the engines. The variation in this quantity can cause variations in the air and fuel mass ratio, increasing the levels of polluting gases due to the occurrence of incomplete reactions (MELO, 2012).

The increase in moisture increases the propensity for the hydrolytic breakdown of the oil, raising the level of free fatty acids, as well as the rancid flavor (TIMILSENA et al., 2017). Moisture is one of the main parameters for marketing the product. High levels of moisture and sedimented products present



in biofuels can accelerate the deterioration processes, saturate the filtering systems and impair the combustion reactions. The analysis of the moisture content in the biodiesel produced with oil extracted from the almond showed a value below that recommended in the legislation, due to the previous drying process of the oil, thus guaranteeing less interference in the reaction and higher quality of the final product.

## CONCLUSION

The research carried out characterized biofuel generated from transesterification by basic catalysis of macauba almond oil. The biofuel produced meets the requirements of the ANP, demonstrating the viability of production. Through the physical-chemical parameters analyzed, it was possible to verify that the biofuel produced presented satisfactory quality.



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# DETERMINATION OF THE VOLUMETRIC EXPANSION COEFFICIENT OF PINEAPPLE AND WATERMELON PULPS

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## ABSTRACT

*The knowledge of the volumetric expansion coefficient ( $\beta$ ) allows to study the variation in density in response to a change in temperature under constant pressure, the expansion behavior, to evaluate the impacts on the volumetric measurement system due to the temperature variation and to design equipment and accessories, considering possible dilations resulting from strong temperature changes. Thus, based on the density data as a function of temperature, it is possible to determine the coefficient of thermal expansion. In fruit processing, the adequate dimensioning of the equipment represents excessive profit in terms of savings and energy. The objective of this work was to determine the volumetric expansion coefficient of pineapple and watermelon pulps as a function of specific masses and temperature variation from 10°C to 60°C, in concentrations of 14.2 °Brix and 8.5 °Brix respectively. The study was carried out at the fruit and vegetable processing unit and at the General Chemistry Laboratory of the Federal Institute of Education, Science and Technology of Tocantins - IFTO Campus Paraíso do Tocantins - TO, between February to April 2019. Ripe pineapples and watermelons were acquired in local shops, covered fair, in the city of*

*Paraíso do Tocantins. The fruits were sanitized and submitted to a pulp extraction process using a multiprocessor. The soluble solids (°Brix) of the pulps were obtained through direct reading with the aid of a portable refractometer, with a scale from 0 to 32 °Brix, properly calibrated and adjusted to 20 °C with distilled water. For the analysis of the specific mass of the pulps, pycnometers previously calibrated with distilled water were used, at temperatures of 10°C, 20°C, 30°C, 40°C, 50°C and 60°C. The temperatures of the samples controlled were by heating and cooling baths and by thermometers. The specific mass calculated was using mathematical equations that established the relationship between pulp mass and pycnometer volume. The volumetric expansion coefficients were performed using the  $\ln \rho_0/\rho = \beta (T-T_0)$  ratio. The values of the specific masses of the pulps decreased with increasing temperature and showed determination coefficients greater than 0.90, showing a good fit.*

### Keywords:

*Linear Regression, Estimation, Activation Energy.*





## INTRODUCTION

**B**razilian fruit production stands out nationally and internationally, due to the availability of the production area, irrigation and favorable climate (EDITORA GAZETA, 2015).

The acquisition of a product with simplicity of storage and preservation of its organoleptic properties, due to its high rate of perishability, makes the processing of frozen fruit pulps an important segment of the production chain, favoring the full use of fruits (SANTOS; BARROS, 2012; SANTOS et al., 2014; SANTOS; FIGUEIREDO NETO; DONZELI, 2016).

Fruit pulp is widely used in the food products industry, such as natural juice, ice cream, dairy, beverage and other industries (MATTA et al, 2005; COSTA, CARDOSO, SILVA, 2013). The processing of fruit pulps is a practice carried out in the agribusiness aimed at increasing the economic value of the fruit, preventing waste and minimizing losses that may occur during the marketing of the product in nature (NASCIMENTO et al., 2013).

During processing, heat transfer, heating, cooling and freezing operations are applied. Knowledge of the thermo physical properties of fruit pulps is necessary for operations that include heat transfer with an interest in determining the time and amount of energy involved during thermal processes (PEREIRA, 2017).

The thermal expansion coefficient ( $\hat{\alpha}$ ), specific gravity, heat capacity and thermal conductivity are thermal properties, as they are related to the reaction of materials due to the application of heat (CALLISTER, 2008). The expansion coefficient provides a measure of the density variation in response to a change in temperature, under a condition of constant pressure (INCROPERA; DEWITT, 2008).

The volumetric expansion coefficient ( $\hat{\alpha}$ ) is an important thermo physical property for designing, dimensioning equipment and processes that involve heat transfer (MATTOS, MEDEROS, 2008). The knowledge of ( $\hat{\alpha}$ ) allows to study the variation in density in response to a change in temperature under constant pressure, the expansion behavior, to evaluate the impacts on the volumetric measurement system due to the temperature variation and to design equipment and accessories, considering possible expansions resulting from strong changes in temperature (INCROPERA, DEWITT, 2008; CANCIAM, 2014).

Thus, based on the data of density as a function of temperature, it is possible to determine the coefficient of thermal expansion (SANTOS; VIEIRA, 2010). In fruit processing, the adequate dimensioning of the equipment represents excessive profit in terms of savings and energy. The production, transport and storage of fruit pulps involve important thermo physical properties for the correct dimensioning of the equipment destined for these operations (LIMA, 2003).



It is essential to know the behavior of the specific mass of the pulps in the processing conditions because it changes during the transfer of heat and mass during the processing of the food (BOLZAN, SOUZA, 2007). During the pulp processing, processes of heating, pasteurization, concentration and the use of low temperatures used are to preserve the quality of these products (LIMA, 2003). The prediction of the behavior of the specific mass under different temperatures through polynomial models is a viable alternative for determining the expansion coefficient of the analyzed pulps (ALVES et al. 2018).

The mathematical models for predicting the thermo physical properties represent an adequacy to expand the efficiency of heat treatments in food processing (EGEA et al., 2015). The objective of this work was the experimental determination of the thermal expansion coefficient ( $\hat{\alpha}$ ) of the whole pulp of pineapple and watermelon at different temperatures from the linear regression analysis of the experimental data using polynomial models of the specific mass as a function of temperature.

## METHODOLOGY

This work was carried out at the fruit and vegetable processing unit and at the General Chemistry Laboratory of the Federal Institute of Education, Science and Technology of Tocantins - IFTO Paraíso do Tocantins - TO campus, between the months of February to April 2019. Acquired in local shops, covered fair, in the city of Paraíso do Tocantins, ripe pineapples and watermelons. The fruits were transported in thermal boxes, to the fruit, vegetable processing unit of the IFTO Paraíso do Tocantins campus, where they underwent a selection process regarding the state of maturity, and integrity with the elimination of those that apparently were damaged or appear spots or injuries. The selected fruits previously cleaned were to remove dirt using water and detergent. After the cleaning process, they were submitted to hygiene with a 100-ppm sodium hypochlorite solution. After 30 minutes, the fruits were removed from the sanitizing solution and subjected to a drying process. With the selected fruits clean and dry, the pineapple and watermelon pulps were extracted using a multiprocessor.

The pineapple and watermelon pulps produced were packaged in low-density plastic bags and stored under refrigeration at 10 ° C until the time of analysis. The soluble solids (°Brix) of the pulps were obtained through direct reading with the aid of a portable refractometer, with a scale from 0 to 32 °Brix, properly calibrated and adjusted to 20 ° C with distilled water.

For the analysis of the specific mass of the pulps, pycnometers previously calibrated with distilled water were used, at temperatures of 10 ° C, 20 ° C, 30 ° C, 40 ° C, 50 ° C and 60 ° C, weighed in an analytical balance. The temperatures of the samples were controlled by heating and cooling baths and by thermometers. The specific mass calculated was using equations (Equation 1), which establishes a relationship between pulp mass and pycnometer volume.

**Equation 1.** Specific mass

Where:  $\rho$  - Product specific mass (kg / m<sup>3</sup>);

$v$  - Volume of the pycnometer (m<sup>3</sup>);

$m$  - Product mass (kg)

$$\ln(\rho) = \beta (T - T_0)$$

For the determination of the thermal expansion coefficient, equation 2 was used, following the same methodology as Canciam (2012):

**Equation 2.** Determination of the expansion coefficient

Where:  $\rho_0$  - specific mass of the pulp at the initial temperature  $T_0$ .

$\rho$  - Specific gravity of the pulp of the final temperature  $T$ .

$\beta$  - Volumetric expansion coefficient.

$$\ln(\rho) = \beta (T - T_0)$$

This methodology considers that the volumetric expansion coefficient ( $\beta$ ) is numerically equal to the slope of the line obtained by the graph of  $\ln(\rho/\rho_0)$  versus  $(T-T_0)$ .

Where  $\rho$  corresponds to the specific mass at temperature  $T$  and  $\rho_0$  corresponds to the specific mass at initial temperature  $T_0$  (CANCIAM, 2012).

## RESULTS AND DISCUSSION

Tables 01 and 02 show the experimental values of specific masses ( $\rho$ ) as a function of temperatures ( $T$ ), soluble solids content ( $^{\circ}$ Brix), volumetric expansion coefficients ( $\hat{\alpha}$ ) and the correlation coefficient ( $R^2$ ) of the specific mass in function of the temperature of the pineapple and watermelon pulps.



PINEAPPLE						
T °C	$\rho$ (Kg.m <sup>-3</sup> )	(T – T <sub>0</sub> )	$\ln(\rho_0 / \rho)$	°Brix (20 °C)	(°C <sup>-1</sup> )	R <sup>2</sup>
10	1060.79 ± 0.16					
20	1058.64 ± 0.13					
30	1054.40 ± 0.47	50.0	0.01929332	14.20	3.9 x 10 <sup>-4</sup>	0.9987
40	1047.69 ± 0.10					
50	1044.56 ± 0.36					
60	1040.52 ± 0.27					

Table 01. Average values ( $\bar{\rho}$ ) as a function of (T), °Brix, ( $\hat{\alpha}$ ) and (R<sup>2</sup>) of the pineapple pulp.

WATERMELON						
T °C	$\rho$ (Kg.m <sup>-3</sup> )	(T – T°)	$\ln(\rho_0 / \rho)$	°Brix (20° C)	(°C <sup>-1</sup> )	R <sup>2</sup>
10	1033.63 ± 0.13					
20	1029.27 ± 0.17					
30	1024.51 ± 0.19	50	0.02864670	8.50	5.7 x 10 <sup>-4</sup>	0.9962
40	1017.39 ± 0.11					
50	1010.51 ± 0.10					
60	1004.44 ± 0.42					

Table 02. Average values ( $\bar{\rho}$ ) as a function of (T), °Brix, ( $\hat{\alpha}$ ) and (R<sup>2</sup>) of the watermelon pulp.

The pineapple pulp showed specific masses higher than the specific masses of watermelon pulp at the same temperatures. This increase related is to the dissolved solids content of the pineapple pulp, 14.2 °Brix, being greater than, 8.5 °Brix, of the watermelon pulp (ALVES et al., 2018). The average values of the specific masses obtained for the pineapple pulp and watermelon pulp decreased with increasing temperature. Nobrega et al. (2019) obtained experimental values for the specific mass of watermelon at temperatures from 10 to 50 °C, varying between 1033.56 to 1010.36 kg/m<sup>3</sup> respectively.

Guedes et al. (2010), studying the behavior of the specific mass of watermelon pulp at different temperatures of 10 to 60 °C and concentrations of 8, 17, 26 and 35 °Brix, observed that the specific mass increases as the concentration of solids occurs soluble and decreases with rising temperature. Alves et al. (2018), researching the behavior of the specific mass of pineapple pulps as a function of temperature, found the values of the specific masses of pineapple pulps ranging from 1060.31 to 1044.32 kg/m<sup>3</sup> at temperatures of 10 to 50 °C. Oliveira et al. (2018) observed during studies on pineapple pulp that the specific mass tended to decrease when there was an increase in temperature in their experiment. According to Mercali et al. (2011), Diniz et al. (2014) and Alves et al. (2018), the



specific mass values of pineapple and watermelon pulps decrease with increasing temperature, probably due to the volumetric expansion of the fluid caused by the reduction of the intermolecular force connection.

The volumetric expansion coefficient ( $\beta$ ) measures the relative change in volume due to the change in temperature at constant pressure. Canciam (2012a), studying the correlation between the volumetric expansion coefficient and the total solids content for pineapple juice between 17.4 and 85.8 °C, with a total solids content of 11°Brix, found the value of  $3.3930 \times 10^{-4} \text{ } ^\circ\text{C}^{-1}$  for the volumetric expansion coefficient. In the literature, no experimental values found were for the volumetric expansion coefficient of pulp or watermelon juice. The coefficient of volumetric expansion of a material being equal to  $1.0 \times 10^{-2} \text{ } ^\circ\text{C}^{-1}$  means that the increase of 1degree results in an increase in volume by 1% (NETZ; ORTEGA, 2008).

The ( $\hat{\alpha}$ ) of pineapple and watermelon pulps showed values equal to  $3.9 \times 10^{-4} \text{ } ^\circ\text{C}^{-1}$  and  $5.7 \times 10^{-4} \text{ } ^\circ\text{C}^{-1}$ , respectively, increasing 0.039% in the volume of pineapple pulp. In addition, 0.057% in the volume of the watermelon pulp with each increase of 1°C in the temperature (NETZ and ORTEGA, 2008). When compared to water ( $\hat{\alpha}$ ) equal to  $2.07 \times 10^{-4} \text{ } ^\circ\text{C}^{-1}$ ), the increases in pineapple and watermelon pulp volumes are respectively 1.88 and 2.75 times greater than that of water (CABRAL and LAGO, 2002). According to Santos and Vieira (2010), it is possible to assess the impacts on the volumetric measurement system from the knowledge of the thermal expansion coefficient of the substance. Considering a volume of 1000 liters of pulp, with a temperature variation of 10°C, the volume of pineapple pulps with 14.2 °Brix increases 3.9 liters and the watermelon with 8.5 °Brix increases 5.7 liters. The volumetric expansion of the fluid generated by the increase in temperature is an effect of the increase of the internal energy causing a greater amplitude of the molecular vibrations and distance between the molecular constituents (CALCIAM, 2017; FRADE, 2010).

The volumetric expansion is directly proportional to the chemical composition of the different liquid substances (JERÔNIMO et al., 2012). Substances that have strong chemical bonds have small values for the distance between chemical species. This is because thermal expansion is related to the asymmetric variation of the energy of connection with the distance between chemical species (CALCIAM, 2017). During heating, there is an increase in the frequency of vibration and distance between chemical species, due to the increase in repulsion forces and a decrease in the forces of attraction (SANTOS; VIEIRA, 2010). The pineapple pulp contains 90.5% water, 6.7% sugars (glucose, fructose and sucrose) and the rest in minerals, vitamins and proteins (MORI, 1998). The watermelon pulp contains 94% water, 5% carbohydrates and 1% distributed in minerals and vitamins (FAO, 2007). Peruzzo and Canto (2010) comment that glucose, fructose and sucrose molecules have hydroxyl groups in their structure. Water is a polar molecule, which interacts between its molecules by hydrogen bonding (BROWN; HOLME, 2009; CALCIAM, 2017).

The formation of hydrogen bonds between the hydroxyls of glucose, fructose and sucrose molecules and water molecules ensures dissolution of these sugars in water and formation of dipole-dipole intermolecular forces between the molecules of these sugars and water (CALCIAM, 2017). Intermolecular forces of the dipole-dipole type are characteristic of polar molecules. The molecules



may have a permanent electric dipole. Due to some distortion in the distribution of electrical charges, one side of the molecule is slightly more "positive and the other slightly more "negative "(CALCIAM, 2017). The tendency of these molecules is to align and interact with each other by electrostatic attraction between the opposite dipoles (CALCIAM, 2017; BROWN; HOLME, 2009).

Hydrogen bonds, when compared with intermolecular forces of the dipole-dipole type, are stronger (BROWN; HOLME, 2009; CALCIAM, 2017). Perhaps for this reason, the volumetric expansion coefficient of pineapple pulp and watermelon pulp is greater than the volumetric expansion coefficient of water (CALCIAM, 2017). According to Toledo and Ovalle (1985), the correlation coefficient ( $R^2$ ) measures the intensity of the linear relationship between the values of the dependent and independent variables in a sample, thus evaluating the quality of the fit. The closer the value of  $R^2$  is to 1, the better the quality of fit of the function to the points of the scatter diagram (CALCIAM, 2017). Figures 01 and 02 illustrate the graphs of  $\ln(\bar{n}/\bar{r}_0)$  versus  $(T-T_0)$  of the correlation coefficients of the pineapple and watermelon pulps, based on the data in tables 01 for the pineapple pulp and 02 for the pulp of watermelon.

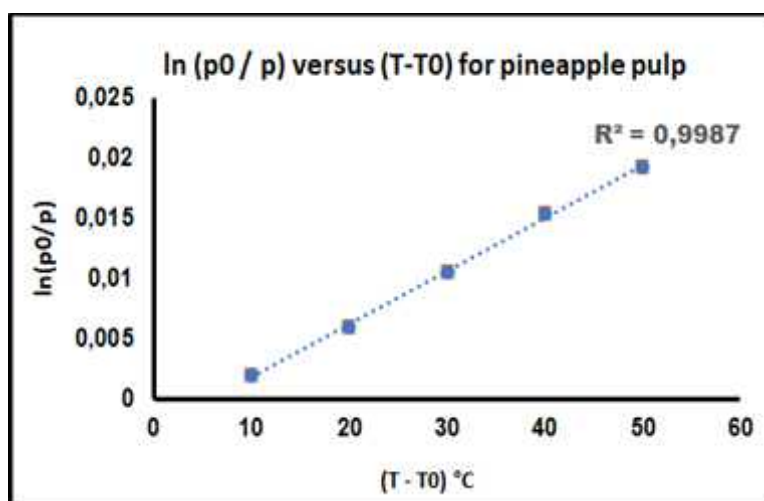


Figure 01. Graph of  $\ln(p_0/p)$  versus  $(T - T_0)$  for pineapple pulp

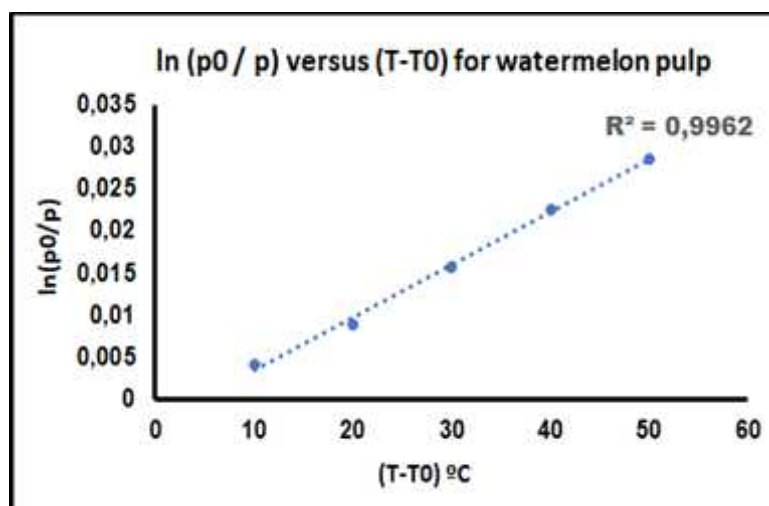


Figure 02. Graph of  $\ln(p_0/p)$  versus  $(T - T_0)$  for watermelon pulp





Lira (2004) states that a linear correlation classified is as very strong when the modules of the values of the correlation coefficient are between 0.90 and 1.0. Thus, according to the author's classification, pineapple and watermelon pulps showed a very strong correlation.

## CONCLUSION

The values of the specific mass of the pulps analyzed decreased with increasing temperature, varying from  $1060.79 \pm 0.16 \text{ kg.m}^{-3}$  to  $1040.52 \pm 0.27 \text{ kg.m}^{-3}$  for pineapple pulp and  $1033.63 \pm 0.13 \text{ kg.m}^{-3}$  to  $1004.44 \pm 0.42 \text{ kg.m}^{-3}$  for watermelon pulp being consistent with the behavior reported for fruit pulp in the literature. The pineapple and watermelon pulps showed volumetric expansion coefficients equal to  $3.9 \times 10^{-4} \text{ }^{\circ}\text{C}^{-1}$  and  $5.7 \times 10^{-4} \text{ }^{\circ}\text{C}^{-1}$  respectively. The values found for the determination coefficients showed a very strong linear correlation.



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# EFFECT OF MICROBIAL TRANSGLUTAMINASE ON THE PROCESSING OF MINAS FRESCAL CHEESE WITH REDUCED SODIUM CONTENT PRODUCED WITH GOAT MILK

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## ABSTRACT

*Goat milk is considered one of the most complete foods since it holds several elements which are important for human nutrition, and necessary for the constitution of tissues and blood. This study evaluated the effect of two concentrations of microbial transglutaminase (TGM: 0.95 and 1.89 IU.g<sup>-1</sup>) and three sodium chloride levels (NaCl: 100%, 70% and 40%) on the yield and in physicochemical characteristics of "Minas Frescal" cheese. The addition of TGM resulted in an increased yield of cheese when compared to the control, being higher when using the highest concentration of enzyme. It was observed the*

*effect of TGM on the increase of moisture content. By increasing the concentration of TGM, an increase in protein content was also observed. On the other hand, it was observed greater calcium retention (781.25g/100mg) in the cheese processed with TGM compared to the control. The results showed that TGM has potential to be used in the formulation Minas Frescal cheese with low sodium content.*

### **Keywords:**

*Yield; Lactic; Enzyme.*





# INTRODUCTION



The production of goat's milk (*Capra hircus*) is of significant importance for the economy and survival of a large part of the populations in various countries of the world (COSTA et al., 2009).

Demand for quality animal products becomes increasingly targeted by the consumer Market generating the search for the production and processing of increasingly elaborate foods with certification of quality guaranteed. This fact is no different for milk goat, which requires the application of methods of differentiated production and processing so that they are improved products offered (RAYNAL-LJUTOVAC et al., 2008) to demystify goat's milk as little food palatable, leading to its expansion nationwide.

Goat milk is a food of high nutritional value and plays a vital role in human nutrition. It is known to contain proteins, vitamins and fatty acids that possess high biological merit (SAFDAR et al., 2020; XU et al., 2015). Goat milk possesses higher levels of calcium, potassium and phosphorus compared to both human and cow's milk and a substantially higher protein concentration compared to human milk (PARK, 2017). It is hypoallergenic and its small fat globules make it easily digestible (PARK, 2017; TZIBOULA-CLARKE, 2003). Moreover, goat milk contains several bioactive peptides with potent antioxidant capacity (DE GOBBA et al., 2014).

Reducing the salt content in food is currently one of the goals to be achieved by the food industry (RODRIGUES et al., 2016). The microbial transglutaminase enzyme (TGM) has been used in the processing of food products as a promising alternative, as it is able to catalyze acyl radical transfer reactions forming intra and intermolecular cross-links in proteins, mainly through covalent bonds between glutamine and lysine residues (HAN et al., 2008; NONAKA et al., 1989). These reactions promoted by the enzyme create profound changes in the protein matrices of food, resulting in technological benefits (DAMODARAN; AGYARE, 2013).

The objective of the present work was to evaluate the effect of the combined use of microbial TGM and salt reduction in the production of minas frescal cheese under the physical-chemical characteristics and yield.



## METHODOLOGY

### Cheese processing

The research was conducted at the Federal Institute of Tocantins (IFTO) - Campus Paraíso do Tocantins, at the Milk and Derivatives Processing Unit and at the Food Analysis Laboratory. The cheeses were processed from whole pasteurized goat milk (Campus Paraíso do Tocantins, Tocantins, Brazil), calcium chloride 40% solution (Ricanata brand), 85% pure lactic acid (Ricanata brand), sodium chloride (refined, CISNE brand), coagulant (CHR Hansen® brand, produced by *Aspergillus niger* var. *awamori* strain) and TGM enzyme (ACTIVA®YG Ajinomoto Interamericana Ind. e Com. Ltda). The treatments resulting from the combination of TGM and reduced salt content were: QC (100% NaCl), Q4 (40% NaCl), Q7 (70% NaCl), Q4T1 (40% NaCl / 0.95 UI.g-1 TGM), Q4T2 (40% NaCl / 1.89 UI.g-1 TGM), Q7T1 (70% NaCl / 0.95 UI.g-1 TGM), Q7T2 (70% NaCl / 1.89 UI.g-1 TGM). Coagulation was carried out at 36 ° C for 45 min, followed by cutting slowly, obtaining cubes of approximately 1.5 cm side, and left to stand for 3 minutes. The mass was stirred for 20 minutes, slowly and smoothly, in order to provide clot retraction and partial expulsion of the serum. After this period the cheese mass was poured into 250 g polypropylene forms for drainage. After the first 60 minutes, the first turning was performed, repeating the process at least three more times at regular intervals of 30 minutes. Then the cheeses were packed in plastic packaging and stored under refrigeration at 5° C.

### Physicochemical analysis

The proximate composition (moisture, protein and lipids) was determined using official methodologies. The moisture determination followed AOAC (2010), the lipids were quantified by the Gerber method, and the protein according to Kjeldahl, multiplying the nitrogen content by the factor 6.38 (Brazil, 2006). For pH measurements, a digital meter was used by inserting the electrode directly into the samples and the acidity was determined by titration, expressed in g / 100g of lactic acid (AOAC, 2010). The ash determination was made according to AOAC (2010).

### Yield

The yield was calculated according to Andreatta et al. (2009) and followed Equation (1):

$$Yield = \frac{\text{obtained mass cheese (g)}}{\text{volume of used milk (L)}} \quad (1)$$



## Statistical analysis

The data were analyzed by ANOVA considering the salt content (100, 70 and 40% NaCl) and the TGM concentration (0.95 and 1.89 IU.g<sup>-1</sup>) as causes of variation and, to check the difference between the averages, the Tukey test was used, at the 5% significance level. Principal Component Analysis (PCA) was used to group the physical and chemical variables according to similarity. PCA was performed on standardized data to avoid the effect of the different levels of magnitude of the response variables. All statistical analyzes were performed using the software R 3.2.4 (2016) developed by Core Team (2011) R, and the package for exploratory analysis of multivariate data, FactorMineR 1.32.

## RESULTS AND DISCUSSION

### Physical-Chemical Analysis and Performance

The Principal Component Analysis (PCA) provided a simplified interpretation (Figure 1) of the information in Table (1), facilitating the visualization of the response variables and them correlating. The main components CP1 and CP2 explained 78.75% of the total variability of the data, with 48.56% explained by the component first and 30.19% by the component second. The sum of main components I and II ( $\geq 75\%$ ) adequately presented the variability between the samples (ABDI; WILLIAMS, 2010).

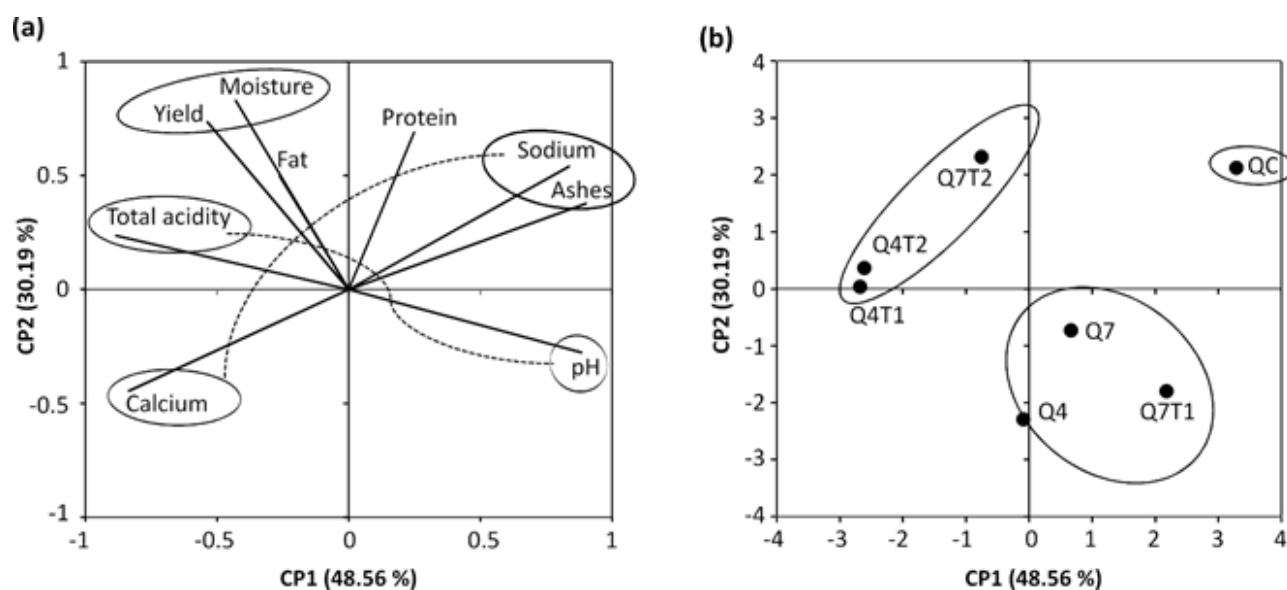


Figure 1- Analysis of main components of the seven samples of Minas Frescal cheese:  
a) representation of the physical-chemical variables and yield; and  
b) representation of the samples.



Table 1 shows the averages obtained in the physical-chemical analyzes and the yield of cheese in the experiments carried out. Significant differences were observed for all parameters ( $p < 0.05$ ).

Physicochemical Parameters	QC	Q4	Q7	Q4T1	Q4T2	Q7T1	Q7T2
Moisture (g/100g)	73.56 <sup>ab</sup> ±0.35	71.97 <sup>bc</sup> ±0.86	71.83 <sup>bc</sup> 0.16	72.84 <sup>ab</sup> ±0.54	74.41 <sup>a</sup> ±0.06	70.5 <sup>c</sup> ±0.38	74.76 <sup>a</sup> ±0.31
Ash (g/100g)	2.55 <sup>a</sup> ±0.05	1.86 <sup>d</sup> ±0.01	2.20 <sup>c</sup> ±0.03	1.84 <sup>d</sup> ±0.02	1.79 <sup>d</sup> ±0.02	2.30 <sup>b</sup> ±0.01	2.18 <sup>c</sup> ±0.01
Protein (g/100g)	17.45 <sup>b</sup> ±0.07	16.98 <sup>b</sup> ±0.17	17.15 <sup>b</sup> ±1.06	19.30 <sup>a</sup> ±0.28	20.65 <sup>a</sup> ±0.21	19.25 <sup>a</sup> ±0.07	20.70 <sup>a</sup> ±0.28
Fat (g/100g)	23.77 <sup>c</sup> ±0.4	23.62 <sup>f</sup> ±0.16	23.82 <sup>c</sup> ±0.70	25.76 <sup>a</sup> ±0.79	24.69 <sup>d</sup> ±0.01	25.26 <sup>b</sup> ±0.38	24.95 <sup>c</sup> ±0.64
Sodium (mg/100g)	558.08 <sup>a</sup> ±0.17	248.05 <sup>f</sup> ±0.14	366.94 <sup>d</sup> ±0.14	240.16 <sup>e</sup> ±0.08	255.91 <sup>c</sup> ±0.15	389.83 <sup>c</sup> ±0.17	396.21 <sup>b</sup> ±0.09
Calcium (mg/100g)	554.446 <sup>e</sup> ±0.15	781.25 <sup>a</sup> ±0.08	654.82 <sup>d</sup> ±0.17	780.18 <sup>b</sup> ±0.04	774.62 <sup>c</sup> ±0.12	614.25 <sup>f</sup> ±0.02	627.76 <sup>c</sup> ±0.10
Yield (g/L)	276.21 <sup>a</sup> ±0.11	265.08 <sup>f</sup> ±0.25	268.88 <sup>c</sup> ±0.50	273.23 <sup>d</sup> ±0.29	293.35 <sup>a</sup> ±0.45	254.01 <sup>e</sup> ±0.78	288.95 <sup>b</sup> ±0.65
pH	6.62 <sup>a</sup> ±0.01	6.61 <sup>ab</sup> ±0.42	6.60 <sup>ab</sup> ±0.35	6.57 <sup>ac</sup> ±0.22	6.61 <sup>a</sup> ±0.28	6.58 <sup>ac</sup> ±0.56	6.52 <sup>c</sup> ±0.65
Total acidity látic (g/100g)	0.11 <sup>d</sup> ±0.01	0.15 <sup>bcd</sup> ±0.01	0.15 <sup>bc</sup> ±0.14	0.18 <sup>b</sup> ±0.24	0.14 <sup>bcd</sup> ±0.12	0.18 <sup>ab</sup> ±0.01	0.20 <sup>a</sup> ±0.02

Table 1- Proximate composition, pH, total acidity and yield of the Minas Frescal cheese prepared with different levels of salt and added with microbial transglutaminase.

It was observed that the humidity was higher than 55% in all cheeses, which was already expected since the Technical Regulation of Identity and Quality of Cheeses (Brazil, 1997) classifies the Minas Frescal cheese as high humidity. However, the samples had a higher moisture content than that normally obtained for the product produced by the traditional process (Felicio et al., 2016). Q4T2 and Q7T2 were the cheeses that obtained the highest moisture values (74.41 and 74.76 g / 100g, respectively), probably due to the ability of microbial transglutaminase to modify the properties of proteins by the formation of the three-dimensional network and covalent bonds between the amino acids glutamine and lysine, helping the water retention capacity in the cheese matrix (Damodaran and Agyare, 2013). Regarding the ash content (mean of 2.20) there was no significant difference ( $p > 0.05$ ) between samples Q4, Q4T2 and Q4T1, as well as there were no differences between samples Q7 and Q7T2. Sant'Ana et al. (2013) reported values between 2.21 and 2.26 g / 100g. These results are in line with those observed by Piccolo (2006) in studies with creamy curd, where the addition of the enzyme transglutaminase did not alter the ash content. Faria (2010) found no significant difference between the samples of milk drink formulated with TGM when compared to the control.

Also, studies conducted with mozzarella cheeses with partial sodium replacement reported no change in chemical composition (moisture, protein, fat and ash) due to the substitution of sodium for potassium (Ayyash et al., 2012). The protein content showed values ranging from 16.98 to 20.7 g / 100g. Values similar to those reported by Felicio et al. (2016) (16.6 - 18.8 g / 100g). Studies with minas frescal cheese added with Lactobacillus showed average values of 13.68 g / 100g (Ribeiro et al., 2009).



In the present study, there was an increase in protein content with an increase in TGM concentration (Table 1), probably due to the greater retention of whey proteins ( $\alpha$ -lactalbumin and  $\beta$ -lactoglobulin) in the dough (Cozzolino et al., 2003).

Regarding the fat content, the values presented significant differences, varying from 23.62% (Q4) to 25.76% (Q4T1), similar to those reported by other authors (Felicio et al., 2016; Gomes et al., 2011). Regarding the sodium levels of the cheese, the values ranged from 558.08 mg / 100 g for QC to 240.16 mg / 100 g for Q4T1. The addition of TGM resulted in an increase in calcium content ranging from 614.25mg / 100g (Q7T1) to 780.18mg / 100g (Q4T1), while the QC showed 554.44 mg / 100 g ( $p < 0.05$ ). Thus, the reduction of sodium led to a decrease in the dissolution of calcium from the para-casein matrix, with a direct effect on the level of colloidal calcium phosphate, resulting in better calcium retention in the cheese matrix (Guinee, 2004). The yield values ranged from 254.01 to 293.35 g / L. The highest values found were for cheeses with higher concentrations of TGM (Q4T2 and Q7T2). The addition of the transglutaminase enzyme at a concentration of 0.06% (1.89 IU.g<sup>-1</sup>) increased the Minas Frescal cheese yield by approximately 6.5% compared to the traditional process. The pH of the samples ranged from 6.52 (Q7T2) to 6.62 (QC) ( $p > 0.05$ ) corroborating with Damodaran and Agyare (2013) who indicated that the pH did not change by using the TGM. The acidity expressed in lactic acid showed values ranging from 0.11 to 0.20 g / 100g. Similar results (0.21) were found by Da Rosa (2004).

## CONCLUSIONS

The highest yield values were found in cheeses with higher concentrations of TGM. The addition of the transglutaminase enzyme at a concentration of 0.06% (1.89 IU.g<sup>-1</sup>) increased the Minas Frescal cheese yield by approximately 6.5% compared to the Minas Frescal cheese produced by the traditional process. However, further studies are needed to better understand the combined use of TGM and the reduction of NaCl in the cheese formulation.



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# ANALYSIS OF PASTY DOCE DE LEITE COMMERCIALIZED IN THE VALLEY OF THE MIDDLE ARAGUAIA REGION - TO

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## ABSTRACT

*Doce de leite is a food with wide production and commercialization in Brazil. Nutritionally, it has a high energy value and a high concentration of proteins, minerals and carbohydrates. The study evaluated the doce de leite sold in the Middle Araguaia TO valley. The analyzes of protein, lipids, moisture, ash and carbohydrates were performed according to the analytical methods recommended by the Adolfo Lutz Institute. The results of the analysis of protein, lipids and ash are within the standards required by Ordinance No. 354/1997, which recommends a minimum: 5g/100g for proteins; 6 to 9g/100g of Lipids;*

*maximum: 30% for moisture and maximum: 2.0 g/100g of ash. The said ordinance does not stipulate reference values for carbohydrates. According to the legislation for doce de leite, the moisture analysis showed non-standard values.*

### Keywords:

*Processing; Milk Derivatives; Centesimal Analysis.*



## INTRODUCTION

Caramel spread is a food with wide production and commercialization in Brazil (MILAGRES et al., 2010). Technical Regulation for Fixing the Identity and Quality of Doce de leite defined it as the product obtained from the cooking of milk added with sucrose, being allowed the use of many other ingredients (BRASIL, 1997).

Still, according to the law, the pasty doce de leite must contain a maximum moisture content of 30% and ash of 2% (w/w). The minimum protein content must be 5.0% (w / w) and the fat content must be between 6.0 and 9.0% (w/w). Sucrose is used to obtain the product and the maximum quantity allowed in the manufacture is 30 kg / 100L of milk. Native or modified starches are allowed in a proportion not exceeding 0.5g / 100mL of milk, as well as mono or disaccharides that replace sucrose in a maximum of 40% (w/w). The enzyme beta-galactosidase (lactase) and sodium bicarbonate can be used as adjuvants, in the amount necessary for good manufacturing practice.

Doce de leite is a product obtained by the concentration of milk added with sucrose, which acquires color, texture and characteristic flavor as a result of non-enzymatic browning reactions, being highly appreciated by consumers (DEMIATE et al., 2001).

Nutritionally, it has a high energy value and a high concentration of proteins, minerals and carbohydrates. Francisquini (2016) states that the doce de leite corresponds to a derivative of great acceptance due to its pleasant sensory characteristic. It is used as an ingredient for the preparation of foods such as cakes, cookies, ice cream, in addition to being consumed directly in food as a dessert or accompanied by bread, toast or cheese (DEMIATE et al., 2001).

The processing of doce de leite is done by many companies, from home-made to large industries, with distribution throughout the country (OLIVEIRA et al., 2012). Most of these companies use customized formulations and production processes, which causes considerable differences in the composition of these products. Regarding production data, Perrone et al. (2012) reported that this information is scarce in Brazil, which makes it impossible to determine the total quantity produced and consumed. However, it is estimated that the production of doce de leite represents approximately 0.6% of the total quantity of products in a dairy.

The quality of the raw material, the type of formulation used and the technological bottlenecks related to the lack of standardization of time and temperature during manufacture have made it difficult to standardize among the brands found in the Brazilian market. Thus, it is imperative to diagnose, study and prioritize how different ingredients, different manufacturing technologies and the intensity of heat treatment influence the quality and uniformity of milk sweets. In view of the above, this study aimed to analyze the physical-chemical composition of pasty doce de leite sold in the Middle Araguaia Valley.



## METHODOLOGY

From January to November 2019, 05 (five) monthly samples of pasty doce de leite brands “A”, “B”, “C”, “D” and “E” were collected within the validity period, at the retail trade, in supermarkets, grocery stores and street markets in the cities of Paraíso do Tocantins, Pugmil and Pium, both located in the Vale do Médio Araguaia Region. At the time of collection, the integrity of the packaging, the storage conditions and the expiration date were observed. Each sample consisted of a 250 g pot of doce de leite. The jars of candy were stored in thermal boxes and transported to the Food Microbiology Laboratory of the Federal Institute of Tocantins - Campus Paraíso do Tocantins.

The samples were analyzed, in triplicate. The physico-chemical parameters were analyzed according to the National Health Surveillance Agency: humidity in an oven at 105 °C until constant weight; ash at 550 °C in muffle until the samples are completely charred; lipids by the Soxhlet method using hexane as a solvent for the extraction of fat from the samples (IAL, 2008); proteins by the Micro-Kjeldahl method, which consists of the determination of total nitrogen, using the factor of 6,38 to convert the result into protein (AOAC, 2010); total carbohydrates by difference [100 - (moisture + ash + lipids + proteins)] and caloric value by multiplying the levels of carbohydrates and proteins in the samples by 4 Kcal.g<sup>-1</sup> and the content of lipids by 9 Kcal.g<sup>-1</sup> (BRASIL, 2005).

The data were submitted to ANOVA analysis of variance and the comparison of means was done by the Tukey test at 5% probability. The means of the parameters were compared with the specific legislation and with values referenced in the literature. The results of the analyzes were compared with the standards required by Ordinance No. 354, of September 4, 1997, from the Ministry of Agriculture, Livestock and Supply (BRASIL, 1997).

## RESULTS AND DISCUSSION

Table 1 expresses the results referring to the average values for the physical-chemical analyzes of doce de leite marketed in the Middle Araguaia Valley, comparing them with the current doce de leite legislation, Ordinance No. 354, of September 4, 1997 (BRASIL, 1997).

Parameters	A	B	C	D	E	Ordinance N <sup>o</sup> . 354 MAPA
Carbohydrates (g/100g)	56.02 <sup>a</sup> ±0.04	63.98 <sup>a</sup> ±0.03	71.12 <sup>a</sup> ±0.03	61.30 <sup>a</sup> ±0.02	66.95±0.01	-----
Ash (g/100g)	1.63 <sup>a</sup> ±0.02	1.57 <sup>a</sup> ±0.01	1.62 <sup>a</sup> ±0.03	1.70 <sup>a</sup> ±0.05	1.53 <sup>a</sup> ±0.07	Max. 2.0 g/100g
Lipids (g/100g)	7.51 <sup>a</sup> ±0.22	7.32 <sup>a</sup> ±0.21	7.74 <sup>b</sup> ±0.22	7.51 <sup>a</sup> ±0.23	7.47 <sup>a</sup> ±0.21	6 a 9 g/100 g
Proteins (g/100g)	5.38 <sup>a</sup> ±0.02	5.27 <sup>b</sup> ±0.01	5.64 <sup>a</sup> ±0.05	5.45 <sup>a</sup> ±0.04	5.53 <sup>a</sup> ±0.2	Min 5 g/100
Moisture (%)	33.70 <sup>a</sup> ±0.17	33.56 <sup>a</sup> ±0.20	33.63 <sup>a</sup> ±0.18	33.73 <sup>c</sup> ±0.15	33.43 <sup>b</sup> ±0.21	Max. 30%

Averages followed by the same letter, on the line do not differ statistically at 5% probability by the Tukey test.

Table 01. Results referring to the average values for the physical-chemical analyzes of doce de leite sold in the Middle Araguaia Valley



The results of the analysis of carbohydrates, ashes, lipids, proteins and moisture are in accordance with the standards required by Ordinance No. 354/1997. Doce de leite showed carbohydrate levels ranging from  $56.02 \pm 0.02\text{g}/100\text{g}$  to  $71.12 \pm 0.03\text{g}/100\text{g}$ . Oliveira et al. (2010), when characterizing commercial doce de leite, found carbohydrate values between  $44.77\text{g}/100\text{g}$  to  $68.98\text{g}/100\text{g}$ . Silva (2016) found values for carbohydrates that ranged from  $57.10\text{g}/100\text{g}$  to  $63.87\text{g}/100\text{g}$ . Brazilian legislation (BRASIL, 1997) does not establish carbohydrate analysis as a mandatory requirement.

The ash contents varied between  $1.53 \pm 0.07\text{g}/100\text{g}$  to  $1.70 \pm 0.05\text{g}/100\text{g}$ . Madrona et al. (2009) developed doce de leite with different proportions of cheese whey and verified ash contents ranging from  $1.31\text{g}/100\text{g}$  to  $2.00\text{g}/100\text{g}$ . Martins et al. (2015) found a variation of 1.50 to  $1.56\text{g}/100\text{g}$  of ash in the samples of doce de leite analyzed. Demiate et al. (2001), states that low ash values may indicate that the products were obtained with little milk or other dairy raw materials. As the mineral content is relatively constant in the raw material, its quantification is an important indicator of the presence of milk in the doce de leite.

Lipids are important in quality control, as they can affect the yield and texture of doce de leite. For this reason, standardized milks must be used in the processing of this dairy derivative, in order to obtain products in compliance with legal specifications. Martins et al. (2015), studying storage of doce de leite, found a variation from 7.1 to  $7.9\text{g}/100\text{g}$  for lipids. Oliveira et al. (2010) reported, for the lipid parameter, 100% failure of the samples of milk sweets sold in Lavras-MG.

Doce de leite had protein contents between  $5.27 \pm 0.01\text{g}/100\text{g}$  to  $5.64 \pm 0.05\text{g}/100\text{g}$ . All samples with protein content above 5%, which is the minimum allowed by law (BRASIL, 1997). Oliveira et al. (2010) also found all samples in compliance with the legislation, with protein contents ranging from  $8.88\text{g}/100\text{g}$  to  $10.49\text{g}/100\text{g}$ . Pieretti et al. (2012) also analyzing pasty doce de leite reported values from  $5.4\text{g}/100\text{g}$  to  $5.9\text{g}/100\text{g}$  for proteins. However, the addition of whey may contribute to some difference in this proportionality, as well as the addition of starch. Feihmann et al. (2006) found a value of  $10.2\text{g}/100\text{g}$  of protein in pasty doce de leite. The authors explained that this value found was due to the maximum temperature used of  $50^\circ\text{C}$ , which promoted less protein denaturation, since in the traditional process, the doce de leite is cooked in open pots and submitted to temperatures above  $100^\circ\text{C}$ . Heat treatment often causes unwanted changes in food, such as changes in taste and loss of functional and nutritional characteristics. All aniseed samples showed moisture above the maximum value established by current legislation. Demiate, Konkel, Pedroso (2001), when studying the chemical composition of several brands of pasty doce de leite, obtained values of moisture between  $22\text{g}/100\text{g}$  and  $32\text{g}/100\text{g}$ , values.

The relatively low moisture of this dairy derivative can influence its microbiological quality (PIERETTI et al., 2012), however, it can also facilitate the appearance of sandiness, a sensory perceived defect in these products. Guimarães et al. (2012). developed doce de leite with coffee and also found all samples with less than 30% moisture, for a variation from 12.45% to 21.70%, already Oliveira et al. (2010) analyzed chemically doce de leite marketed in Lavras-MG and found that the samples showed moisture ranging from 15.57% to 39.03%, with 50% of the analyzed doce de leite failing. Francisquini et al. (2016) also analyzing doce de leite found a variation of 25.40% to 44.87% of moisture.



Passos et al. (2013) when evaluating the quality of pasty doce de leite commercial brands marketed in the Alto Paranaíba / MG region, observed that the moisture content of the samples varied between 20.56% to 34.57%; ash 0.91 g/100g to 3.18g/100g and protein content 2.14 g/100g to 5.86g/100g. The results obtained by the referred authors differ from those found in this research.

## CONCLUSIONS

According to the legislation for doce de leite , the samples did not comply with Ordinance No. 354, of September 4, 1997, for the moisture parameter. For the contents of proteins, lipids, ash and carbohydrates, all samples showed values within the quality standards required by the aforementioned legislation.





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# PHYSICAL-CHEMICAL AND MICROBIOLOGICAL CHARACTERISTICS OF RICOTTAS COMMERCIALIZED IN THE CITY OF PARAÍSO DO TOCANTINS

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## ABSTRACT

*Ricotta cheese is high in moisture and nutrients. These conditions, combined with pH, allow the multiplication of microorganisms that can cause food poisoning and risk to public health. The research analyzed the physical-chemical and microbiological composition of the ricotta commercialized in the city of Paraíso do Tocantins. The physical-chemical analyzes of titratable acidity, ashes, carbohydrates, pH, moisture, fats and proteins used the Official Analytical methods described in INA 68 of December 12, 2006, of MAPA. For the evaluation of total and thermotolerant coliforms and research for*

*Salmonella spp, the methods described in Normative Instruction Nº. 62, of August 26, 2003, from the Ministry of Agriculture, Livestock and Supply were used. The results obtained by the analysis of the ricotta brands show that the contents of moisture, fat, thermotolerant coliforms (45°) and Salmonella spp. are in accordance with the parameters established in the legislation.*

### Keywords:

*Cheese; Physical-Chemical Control; Technology.*



## INTRODUCTION

Cheese whey is considered the main by-product of the dairy industry and has high nutritional value, conferred by the presence of proteins with a high content of essential amino acids, relevant functional properties, and can be used for the production of ricotta (CARVALHO et al, 2015; RICHARDS, 2002; LEITE, 2006).

Ricotta is a cheese of Italian origin that has expanded all over the world, characterized by high moisture and short shelf life (MANCUSO et al., 2014). In Brazil, ricotta is also known as albumin cheese because it consists of lactoalbumin and lactoglobulin. These proteins are the main components of serum and essential for improving the immune system (PICCOLI et al., 2005).

Cheese whey proteins are easily denatured and precipitated by heat, under the influence of acidification, which constitutes the basic principle of making ricotta (ESPER, 2006; HARAGUCHI et al., 2006). The ricotta cheese is produced by heating the cheese whey together with bovine milk (up to 20% of the total volume) in an acidic medium (acetic acid or citric acid) at 80 to 90 °C and subsequent rest of approximately 25 minutes to promote the precipitation of the whey and milk proteins (SANSONETTI et al, 2009). This heat treatment results in the denaturation of the residual protein, mainly albumin and globulin, and keeps the fat globules remaining from the cheese processing (GAMMARIELLO et al., 2009; MANCUSO et al., 2014).

Then, it is hung and packed at temperatures between 2.0 and 4.0 °C (MORAIS et al., 2003). According to Esper (2006), when fresh, the ricotta has 72% of dry matter, where 8 to 12% refer to proteins and 3% to lactose, with a moisture content of not less than 55% and an average yield of 4.0 to 5.0%. Due to the addition of milk in its production (up to 20% of the total volume), allowin for its greater yield, the content of total fats in its mass can rise between 1 and 2%. This fresh type cheese, which, due to its low fat content, absence of salt and easy digestion, has become one of the most consumed foods in diets (RIBEIRO et al., 2005).

Cheese production is a beneficial alternative from an ecological point of view, since in its manufacture, serums from other cheeses (minas, standard mines and mozzarella) are reused, representing advantages from the economic point of view, due to the reduction of expenses in the waste treatment and the optimized use of the raw material (MORAIS et al., 2003). Ricotta is a product whose conservation is limited due to high moisture content (70 to 73%) and availability of nutrients, such as mineral salts and lactose (MAIA et al., 2004). These conditions, combined with the generally high pH, favor the multiplication of contaminating microorganisms, whether these are deteriorating or pathogenic, which can represent a risk to public health, causing food poisoning and toxinfection (PINTADO, MACEDO, MALCATA, 2001). This work aims to analyze the physical-chemical and microbiological composition of ricotta commercialized in the city of Paraíso do Tocantins.



## METHODOLOGY

The ricotta samples were acquired monthly from August to December 2019, in supermarkets located in the city of Paraíso do Tocantins, State of Tocantins, totaling 20 samples analyzed. Each ricotta brand represents a treatment, from a set of samples that were ordered in letters A, B, C and D. The experimental design used was entirely casualized (DIC), with four treatments and seven repetitions.

At the time of collection, the integrity of the packaging, storage conditions and expiration date were observed. All samples were analyzed and inspected by the Federal Inspection Service (SIF). After collection, the samples were placed in isothermal boxes with recyclable ice and sent to the Food Laboratory of the Federal Institute of Tocantins IFTO Campus Paraíso do Tocantins for the analysis of physical-chemical and microbiological tests.

The physical-chemical analyzes of titratable acidity, ashes (Fixed Mineral Residue), carbohydrates (difference [100 - (moisture + ash + fats + proteins)]), hydrogen potential pH, moisture, fats and proteins used the Official Analytical methods described in IN ° 68 of December 12, 2006, from the Ministry of Agriculture, Livestock and Supply (BRASIL, 2006). All analyzes were performed in triplicate.

The ricotta were subjected to analyzes of total coliforms (36 °C) and thermotolerants (45 °C) and *Salmonella* spp. To assess the microbiological quality of the ricotta, the methods described in Normative Instruction No. 62, of August 26, 2003, from the Ministry of Agriculture, Livestock and Supply (BRASIL, 2003) used were.

The computer program Assistat version 7.7 beta was used to apply the Analysis of Variance (ANOVA) and comparison of means by the Tukey test at 95% probability (SILVA; AZEVEDO, 2016).

## RESULTS AND DISCUSSION

Ricotta cheese does not have a specific Technical Identity and Quality Regulation, which makes it difficult to standardize the manufacturing technology and the microbiological and physical-chemical inspection of the final product. The lack of legal standards can be detrimental to the official quality control of these products.

The lack of definition of physical-chemical parameters makes it difficult to interpret the microbiological results established in the Resolution of the Collegiate Board (RDC) No. 12 of January 2, 2001, of the National Health Surveillance Agency (ANVISA). Table 01 shows the microbiological and physical-chemical results of 4 different brands of ricotta cheese sold in supermarkets in the city of Paraíso do Tocantins TO.



Physicochemical analysis	Ricotta A	Ricotta B	Ricotta C	Ricotta D
Acidity (g lactic acid / 100 g)	0.19 <sup>3</sup> ± 0.02	0.13 <sup>2</sup> ± 0.08	0.09 <sup>1</sup> ± 0.41	0.21 <sup>4</sup> ± 0.58
Carbohydrate (g / 100 g)	5.22 <sup>10</sup> ± 0.08	5.41 <sup>12</sup> ± 0.20	5.61 <sup>14</sup> ± 0.31	5.23 <sup>11</sup> ± 0.11
Ash (g/100 g)	2.01 <sup>7</sup> ± 0.08	2.34 <sup>8</sup> ± 0.13	1.80 <sup>5</sup> ± 0.16	1.95 <sup>6</sup> ± 0.14
Fat (g/100 g)	9.56 <sup>17</sup> ± 0.34	10.07 <sup>20</sup> ± 0.25	9.87 <sup>19</sup> ± 0.28	9.69 <sup>18</sup> ± 0.44
Hydrogen potential pH	5.41 <sup>13</sup> ± 0.04	5.76 <sup>15</sup> ± 0.03	5.95 <sup>16</sup> ± 0.02	5.12 <sup>9</sup> ± 0.06
Protein (g/100 g)	12.85 <sup>23</sup> ±		12.76 <sup>22</sup> ±	
	1.23	12.63 <sup>21</sup> ± 1.34	0.94	13.02 <sup>24</sup> ± 2.15
Moisture (g/100 g)	70.36 <sup>28</sup> ± 0.48	69.56 <sup>25</sup> ± 0.63	0.55	70.11 <sup>27</sup> ± 0.39
Total coliforms at 35 °C (NMP/g)	< 1 x 10 <sup>1</sup>	< 3 x 10 <sup>1</sup>	< 2 x 10 <sup>1</sup>	< 1 x 10 <sup>1</sup>
Thermotolerants coliforms 45 ° C (NMP / g)	< 1 x 10 <sup>1</sup>	< 1 x 10 <sup>1</sup>	< 1 x 10 <sup>1</sup>	< 2x 10 <sup>1</sup>
Research of <i>Salmonella</i> spp	Absence	Absence	Absence	Absence

Averages followed by the same number on the lines do not differ by the Tukey Test (p <0.05)

Table 01. Physical-chemical and microbiological results of the analysis of ricotta cheese brands sold in supermarkets in the city of Paraíso do Tocantins TO

There were significant differences at the 5% level (p <0.05) for titratable acidity, ash, carbohydrates, potential pH of hydrogen, moisture, fats and proteins, indicating the existence of heterogeneity between the ricotta marketed in the municipality of Paraíso do Tocantins. This can be justified by the possible variation in the breed of dairy cows, in the process of preparing these products, serum composition, and clotting time. The microbiological results shown in table 01, show a variation of <1 x 10<sup>1</sup> NMP/g to <3, 1 x 10<sup>1</sup> NMP/g in the total coliform count (35 oC), <1 x 10<sup>1</sup> NMP/g to <2 x 10<sup>1</sup> NMP/g for thermotolerant coliforms (45 oC) and absence of *Salmonella*. These results are in accordance with Resolution - RDC No. 12/2001 (BRAZIL, 2001) which determines a limit value of 5x10<sup>2</sup> NMP/g for thermotolerant coliforms in cheese with high moisture and absence of *Salmonella*. This legislation does not present parameters for total coliforms (35 oC). Values higher than those found in this study were observed by Hipólito et al (2013), evaluating the hygienic-sanitary quality of ricotta cheese sold in Alfenas MG.

The presence of *Salmonella* sp and large amounts of thermotolerant coliforms represent potential risks to public health. The bacteria in the coliform group serve as indicators of contamination and the presence of these microorganisms in processed foods indicates failures in processing, poorly sanitized utensils and equipment or post-contamination at the time of operation. The pH and titratable acidity generally correlate in an inverse way (CECCHI, 2003). This fact was observed in the present study. Esper (2006) found values for titratable acidity much more varied (0.13% to 1.25%) than those found in the present study. Ricotta had an average carbohydrate content ranging from 5.22% to 5.61%.





The carbohydrate content was calculated by difference according to RDC nº 360 of 12/23/2003. The percentage of ash found in the analyzed brands ranged from 1.80% to 2.34%, this variation range being greater when compared to the percentage of ash found by Lacerda et al. (2011) who obtained a variation of 1.02 and 2.09%. Regarding the content of fats, the ricotta varied from 9.56% to 10.07%, with the levels above the range reported by Detoni and Gonçalves (2011) for ricotta cream being 2.55% to 3, 25%.

Elevated fat values were also reported by Pellegrini et al. (2012) and Silva and Ferreira (2010), who reported ricotta with fat levels between 19.34 to 22.57% and 6.40 to 20.00%, respectively. According to Cruz and Gomes (2001), the addition of 20% milk to the total volume of whey can cause an increase in the fat content by up to 2%. According to Ordinance No. 146 (BRASIL, 1996), ricotta is a type of cheese and can be classified as a lean product, from 10 to 25% fat. The pH values obtained in the present study varied significantly. Pellegrini et al. (2012), in a study on fresh ricotta made from cow's milk, found a protein content of 19.27%, higher than the protein value found in this research. All analyzes showed a high percentage of moisture, ranging from 69.56% to 70.36%.

The results obtained, according to Ordinance No. 146 of the Ministry of Agriculture, Livestock and Supply (BRASIL, 1996), in the present study fall into the classification of cheese of very high moisture, for presenting values above 55%. The values found are in agreement with those described by Esper (2006), which ranged from 58.49% to 77.45% in ricotta marketed in the city of Campinas-SP

## CONCLUSIONS

The results obtained from the analysis of the ricotta brands A, B, C and D marketed in the city of Paraíso do Tocantins show that the contents of moisture, fat, thermotolerant coliforms (45°) and *Salmonella* spp. are in accordance with the parameters established in RDC No. 12 of January 2, 2001 and Ordinance No. 146 of March 7, 1996. It is necessary to disseminate a standardization of the technology for the preparation and microbiological and physical-chemical inspection of these products, through competent inspection agencies, as the lack of legal standards can be detrimental to the official quality control of these products.



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# ASSESSMENT OF PHYSICAL CHEMICAL AND MICROBIOLOGICAL QUALITY OF FROZEN TROPICAL FRUIT PULPS COMMERCIALIZED IN THE REGION OF THE VALLEY OF THE MIDDLE ARAGUAIA - TO

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## ABSTRACT

*The need for guidelines for the elaboration of Identity and Quality Standards for frozen fruit pulps is necessary due to the organoleptic characteristics: color, flavor, aroma and texture and undesirable sanitary quality in some sectors. The research aimed to analyze the physical and chemical parameters of pineapple, acerola, cashew, guava, mango and passion fruit pulps commercialized in Vale do Médio Araguaia - TO. The analyzes performed were soluble solids; Total titratable acidity; Vitamin C and hydrogen potential (pH) and reducing sugars. The analyzes followed the methodologies recommended by the*

*physical-chemical methods of food analysis of the Adolf Lutz Institute (2008), According to standards of identity and quality of fruit pulp recommended in Normative Instruction SDA No. 37 OF 01/10/2018, the pulps of acerola, guava, passion fruit and mango were presented the chemical physical parameters analyzed within the standard required by the legislation, thus, becomes suitable for human consumption.*

### **Keywords:**

*Processing; Quality Standard; Vitamin C.*



# INTRODUCTION



Tropical fruits are important sources of antioxidants, but most of these fruits are quite perishable and their post-harvest losses can be reduced by processing the fruit into pulps (MAIA et al., 2009).

Therefore, fruit processing aims to increase the shelf life of products, add value to the new product, facilitate consumption, especially seasonal ones, by disseminating them to other regions where there are no certain products and taking advantage of surplus production, providing greater profitability in commercialization (SILVA et al., 2010).

The preparation of the pulps must be carried out only with clean and healthy fruits and free any foreign material in its natural composition. Fruit pulps have some general characteristics, such as high water activity, high oxy-reduction potential and low hydrogen potential pH (PEREIRA et al, 2006). In the quality control of fruit pulps, parameters such as pH, titratable acidity, soluble solids, reducing and total sugars, and vitamin C are important for product standardization and analysis of possible changes that occurred during processing and or storage (BENEVIDES et al., 2008).

The freezing of fruit pulp is a conservation method that preserves the characteristics of the fruit and allows its consumption in the off-season (MATTA et al. 2005). In Brazil, the quality of fruit pulps is regulated by Normative Instruction n.1, of January 7, 2000, which determines the Identity and Quality Standards (PIQ). This resolution defines fruit pulp as the unfermented, non-concentrated and undiluted product, obtained from pulpy fruits through an appropriate technological process, with a minimum content of total solids, from the edible part of the fruit (BRASIL, 2000).

The need for guidelines for the development of Identity and Quality Standards (PIQ) for frozen fruit pulps is necessary due to the undesirable quality of some industries (OLIVEIRA et al., 1998). Given the above, the objective of the present study was to evaluate the physical-chemical quality of frozen pineapple, acerola, cashew, guava, mango and passion fruit pulps marketed in the Middle Araguaia Valley Region and to compare the results obtained with the current legislation.

## METHODOLOGY

From February to November 2019, monthly samples of acerola, cashew, guava, mango and passion fruit pulps of brands "A" and "B" were purchased in the city of Paraíso do Tocantins, Pium, Pugmil and Miranorte, both located in the Region of the Valley of the Middle Araguaia. Sample collection consisted of random selection of 100g units. The labels on the pulp packaging indicated an expiration date of 12 months. The experiment was developed in a completely randomized design and all analyzes were performed in triplicate. The samples were packed in thermal boxes and sent for analysis at the Food Analysis laboratory of the Federal Institute of Science, Technology and Education of Tocantins-TO, Campus Paraíso do Tocantins.





For all analyzes, the fruit pulps were thawed, homogenized and were not diluted. The samples were placed in a 100ml beaker, with three replicates for each flavor. The hydrogen potential (pH) was measured by diluting 10 g of the sample in 100 mL of distilled water, using a digital pot, previously calibrated with buffers of (pH) 4.0; 7.0 and 10.0.

The analysis of titratable total acidity (TTA) was performed by diluting approximately 5 grams of the sample in 20 mL of water, accompanied by 5 drops of phenolphthalein. After these procedures, the titration was done with a sodium hydroxide solution (NaOH 0.1 mol / L). The colored samples of guava and acerola had their stains removed by activated carbon before titration with a 0.1 N Sodium hydroxide solution. Soluble Solids (BRIX) was analyzed by Refractometry, a direct reading was performed on an Abbé bench with scale ranging from 0 to 32 °Brix at room temperature (25 °C). The analysis of vitamin C was performed by the potassium iodate method and the reducing sugars (AR) were determined by the sulfuric phenol method.

The physical chemical parameters analyzed followed the methodology indicated by the Adolf Lutz Institute (IAL, 2008) and compared with Normative Instruction SDA No. 37 of 01/10/2018 that establishes the analytical parameters of juice and fruit pulp (BRASIL, 2018). The data were submitted to ANOVA analysis of variance and the comparison of means was done by the Tukey test at 5% probability.

## RESULTS AND DISCUSSION

The results of the physical-chemical analyzes of the fruit pulps analyzed are presented in Table 1. For comparison purposes, the values are shown in Table 2, according to Normative Instruction SDA No. 37 OF 10/01/2018.

Pulp	Brand	AR (g/100g)	ATT (g/100g)	(pH)	°Brix (a 20°C)	Vitamin C (mg/100g)
Pineapple	A	7.83 <sup>ab</sup> ±0.03	0.76 <sup>a</sup> ± 0.01	3.67 <sup>ab</sup> ± 0.11	14.29 <sup>a</sup> ±0.11	13.13 <sup>b</sup> ± 0.10
	B	8.35 <sup>ab</sup> ±0.30	0.77 <sup>a</sup> ± 0.03	3.53 <sup>ab</sup> ± 0.10	14.03 <sup>a</sup> ±0.10	10.2 <sup>b</sup> ± 0.13
Acerola	A	2.26 <sup>c</sup> ±0.02	0.89 <sup>a</sup> ± 0.02	3.45 <sup>ab</sup> ± 0.12	7.92 <sup>c</sup> ± 0.10	830.1 <sup>a</sup> ± 0.05
	B	2.12 <sup>c</sup> ± 0.01	0.83 <sup>a</sup> ± 0.10	3.14 <sup>ab</sup> ± 0.30	7.92 <sup>a</sup> ± 0.20	885.9 <sup>a</sup> ± 0.10
cashew	A	3.17 <sup>b</sup> ±0.10	0.49 <sup>a</sup> ± 0.10	3.85 <sup>a</sup> ± 0.10	10.97 <sup>abc</sup> ± 0.08	79.2 <sup>a</sup> ± 0.10
	B	3.26 <sup>b</sup> ±0.02	0.42 <sup>a</sup> ±0.05	4.34 <sup>a</sup> ± 0.12	10.47 <sup>abc</sup> ± 0.10	71.4 <sup>a</sup> ± 0.08
Guava	A	2.80 <sup>c</sup> ±0.03	0.48 <sup>a</sup> ± 0.04	3.83 <sup>ab</sup> ± 0.08	10.19 <sup>bc</sup> ± 0.13	10.24 <sup>b</sup> ± 0.00
	B	2.55 <sup>c</sup> ±0.10	0.5 <sup>a</sup> ± 0.10	4.16 <sup>ab</sup> ± 0.13	10.06 <sup>bc</sup> ± 0.10	10.93 <sup>b</sup> ± 0.12
Mango	A	2.63 <sup>c</sup> ±0.20	0.58 <sup>a</sup> ± 0.04	3.89 <sup>ab</sup> ±0.10	12.8 <sup>ab</sup> ± 0.16	66.14 <sup>b</sup> ± 0.20
	B	2.47 <sup>c</sup> ±0.16	0.57 <sup>a</sup> ±0.00	3.78 <sup>ab</sup> ± 0.07	12.8 <sup>ab</sup> ± 0.15	66.14 <sup>b</sup> ±0.05
Passion fruit	A	8.88 <sup>ab</sup> ±0.03	2.64 <sup>a</sup> ± 0.011	3.15 <sup>b</sup> ±0.10	11.25 <sup>abc</sup> ± 0.10	8.89 <sup>b</sup> ± 0.23
	B	8.72 <sup>ab</sup> ±0.20	2.57 <sup>a</sup> ±0.10	3.02 <sup>b</sup> ± 0.05	11.75 <sup>abc</sup> ± 0.20	8.65 <sup>b</sup> ±0.21

Table 1. Physical and chemical parameters of frozen fruit pulps sold in the central region of City in Paraíso do Tocantins.



Averages followed by the same letter, in the column do not differ statistically at 5% probability by the Tukey test.

Normative Instruction SDA Nº. 37 OF 10/01/2018 that approves the General Technical Regulation for setting the Identity and Quality Standards PIQ for Fruit Pulp										
Pulp	AR (g/100 g)		ATT (g/100 g)		pH	°Brix, a 20° C		Vit C (mg/100 mg)		
	Mín	Máx	Mín	Máx	Mín	Máx	Mín	Máx	Mín	Máx
Pineapple	-----	15.00	0.30	-----	-----	-----	11.00	-----	-----	21.50
Acerola	4.00	-----	0.80	-----	2.80	-----	5.50	-----	800.0	-----
cashew	-----	-----	0.18	-----	3.80	-----	10.00	-----	80.00	-----
Guava	-----	-----	0.40	-----	3.50	-----	7.00	-----	24.00	-----
Mango	-----	-----	0.30	-----	3.50	-----	11.00	-----	6.10	-----
Passion fruit	-----	-----	2.50	-----	2.70	-----	11.00	-----	-----	-----

Table 2. Identity and Quality Standards for frozen fruit pulps.

Analysis of pH, acidity, soluble solids and vitamin C for pineapple, acerola, cashew, guava, mango, passion fruit and vitamin C pulp presented values within the limits established by legislation. The maximum and minimum parameters for the reducing sugars analyzed in cashew, guava, mango and passion fruit pulps are not established in the aforementioned legislation. The pineapple and acerola pulps presented values for the analysis of reducing sugars within the recommended by legislation. Côrtes et al (2016) researching sugar content in eight different types of fruit found a percentage of 0.58% for acerola, 2.87% for cashew, 1.88% for guava and 1.19% for passion fruit for analysis of reducing sugars (glucose). Lima et al (2015), evaluating the vitamin C content of fruit pulps sold in five municipalities in Alto Sertão da Paraíba, found a variation of  $2.04 \pm 0.18$  for pineapple,  $401.74 \pm 8.38$  mg/100g for acerola,  $2.52 \pm 0.03$ mg/100g for mango,  $6.21 \pm 0.05$  for passion fruit and  $4.35 \pm 0.84$  mg/100g for guava. The results obtained for vitamin C are different from those described by Bueno et al. (2002) and Pinheiro et al. (2006). Comparing the levels of total titratable acidity of the samples with other works in the literature, it is observed that the results obtained for the guava and cashew pulps showed lower values than found by Pinheiro (2006), the same happens for the pineapple and mango pulp which obtained values well below those described by Bueno (2002) and Pinheiro (2006).

The passion fruit pulp obtained a total titratable acidity value similar to that of RAIMUNDO (2009). Dantas et al. (2010), analyzing vitamin C in acerola pulps in the city of Campina Grande-PB, found an average value of 971.51 mg / 100g. As there are no values in the current legislation, the minimum vitamin C for passion fruit pulp made comparisons with works by other authors. In this research, the determination of vitamin C in passion fruit pulp found results ranging from 63.65 0.05 mg / 100g



which are higher than those reported by Gomes et al. (2006). The ATT values expressed in citric acid, in the pulp of guava, mango and passion fruit were in accordance with the standard established by legislation.

The acidity of the fruit is an important parameter in the analysis of its conservation status, since malic, tartaric, citric and pyruvic acids inhibit the growth of certain microorganisms. (CHITARRA; CHITARRA, 2005). All pulps analyzed showed soluble solids that were above the values established by Normative Instruction SDA No. 37 OF 10/01/2018. (BRASIL, 2018).

The content of Total Soluble Solids ( $^{\circ}$ Brix) is used as an indirect measure of the sugar content, as its value increases as they accumulate in the fruit. However, its determination does not represent the exact content of sugars, as other substances also dissolve in the vacuolar sap (vitamins, phenolics, pectins, organic acids), although sugars are the most representative and can constitute up to 85 to 90% of these (CHITARRA; CHITARRA, 2005). Edaphoclimatic factors and organic acids, important components in the formation of different properties of fruits, contribute to pH variation (SANTOS et al., 2014).

Silva et al (2015), analyzing pH in fruit pulp in the Sertão da Paraíba, found values ranging from 1.22 to 4.55. There was a divergence in the pH values of pineapple, acerola, cashew, guava, mango and passion fruit pulps with those described by Silva et al. (2015) and Bueno et al. (2002)

## CONCLUSIONS

According to the data obtained and discussed, it is concluded that the physical-chemical characterization of frozen tropical fruit pulps of pineapple, acerola, cashew, guava, mango and passion fruit meets the requirements of Brazilian legislation and the results obtained are in agreement with those described in the literature. So, it can be said that such a product becomes suitable for human consumption.



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# EVALUATION OF TROPICAL FRUIT NECTARS COMMERCIALIZED IN THE MIDDLE ARAGUAIA REGION - TO

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## ABSTRACT

*The nectar market expanded due to the consumer's lack of time to prepare the juice. It is possible that nectars are at variance with the parameters of current legislation. This possibility has an impact on the health of children and adolescents, who generally appreciate the intake of this type of drink. The present work aims to evaluate the physical-chemical parameters in nectar of acerola, cashew, guava, mango and passion fruit commercialized in the middle Araguaia region - TO and to compare with the current legislation. Determinations of pH, acidity, soluble solids, vitamin C analysis were carried out*

*following the methodology described at Instituto Adolfo Lutz. Analyzes were carried out in triplicates and their results were compared with Normative Instruction No. 12/2003 of MAPA. Acerola, cashew and guava nectars showed total titratable acidity, soluble solids and Vitamin C within the quality standards established by Normative Instruction No. 12, of September 4, 2003 MAPA.*

### **Keywords:**

*Fruits; Consumer; Processing.*





# INTRODUCTION



The term “Drink” refers to any industrialized product, intended for human consumption, in a liquid state marketed in various ways: concentrated, dehydrated, whole, in the form of nectar, with or without added sugar and preservatives, frozen, freeze-dried and with or without recovery of aromas (LIMA, 2011). Nectar processing is a highly important agro-industrial activity, as it increases the product's useful life, economically values the fruit, standardizes quality, avoids waste and thus minimizes costs (ARANTES, 2012).

In Brazil, fruit drinks include juice and nectar, which, according to Brazilian legislation, differ in the amount of fruit pulp that is added in each formulation, with nectar being the drink with the lowest proportion of fruit pulp. (CARNEIRO, 2013; MOURA; FIGUEIRÊDO; QUEIROZ, 2014). Nectar is the product with the minimum amount of declared fruit pulp, which must be at least 30% w/w (mass/mass) for fruits with high acidity or very strong flavor, the minimum pulp content must be 20% (m/m) (BRASIL, 2003). Juices or nectars are consumed by all age groups and are considered healthy and refreshing drinks. They have high acidity and can naturally contain microbial growth inhibitors and additives such as dyes and flavorings (PIMENTEL; PRUDENCIO; RODRIGUES, 2011).

The acceptance and quality of juice or nectar related are to the quality of the raw material used, the relationship between soluble solids and its acidity, as well as the process of obtaining and storing it (LEITÃO, 2007). Currently, the market for nectar has expanded significantly because it is a non-alcoholic beverage and due to the consumer's lack of time to prepare juice from fresh fruits (ARANTES, 2012). The characteristics of the fruits, the practicality of the packaging and the innovation of the product in providing the necessary information contribute to the development of the product (FERRAREZI, 2008).

It is possible that the flavors of nectars of greater availability and accessibility to the public are at odds with the parameters of the current legislation. This possibility has an impact on the health of children and adolescents, who generally appreciate the intake of this type of drink. The present work aims to evaluate the physical-chemical and microbiological parameters in nectars of acerola, cashew, guava, mango and passion fruit marketed in the cities of Barrolândia, Paraíso do Tocantins, Miranorte, Pugmil both located in the middle Araguaia region - TO and compare current legislation.

## METHODOLOGY

The study was developed at the food laboratory of the Federal Institute of Tocantins - IFTO, campus Paraíso do Tocantins. Samples of acerola, cashew, guava, mango and passion fruit nectar were purchased from four (4) different brands “A”, “B”, “C” and “D”, in their original 200 ml packaging, available in local shops in the city of Paraíso - TO, both brands being within their respective validity terms.



Determinations of hydrogen potential - pH were performed with the aid of a digital pH meter, previously calibrated with pH 4.0 buffer solutions; 7.0 and 10.0. Acidity, through titration with 0.1 M sodium hydroxide and alcoholic phenolphthalein solution (1%), the results being expressed in g of citric acid/100g.

Total soluble solids, by refractometry at 20 °C, expressed in degrees (°Brix). Vitamin C analysis was performed using the potassium iodate method. The physical chemical parameters analyzed were followed the methodologies indicated by the Adolf Lutz Institute (IAL, 2008) and were compared with Normative Instruction nº 12, of September 4, 2003, from MAPA. Microbiological analyzes of total and thermotolerant coliforms were performed using the Multiple Tube technique, according to procedures described by the National Health Foundation (FUNASA, 2006) and were compared with Consolidation Ordinance No. 5, of September 28, 2017 (BRAZIL, 2017).

The data submitted were to ANOVA analysis of variance and the comparison of means done was by the Tukey test at 5% probability. The means of the parameters compared were with the specific legislation and with values referenced in the literature. The averages were expressed in mean values  $\pm$  standard deviation and compared with the legislation to verify compliance with the Identity and Quality Standard (PIQ) provided for in Normative Instruction No. 12, of September 4, 2003 (BRASIL, 2003).

## RESULTS AND DISCUSSION

According to the values expressed, in Tables 01 and 02, it be seen can that the results of the physical-chemical analyzes of the samples of nectars are in accordance with the recommendations in Normative Instruction Nº. 12, of September 4, 2003 of MAPA.

Nectar	Mark	Titratable acidity ATT (g citric acid / mL)	Vitamin C (mg ascorbic acid/100 mL <sup>-1</sup> )	hydrogen potential pH	Soluble Solids (°Brix)
Acerola	A	0.31 <sup>A</sup> $\pm$ 0.01	231.23 <sup>C</sup> $\pm$ 0.25	2.97 <sup>A</sup> $\pm$ 0.02	10.50 <sup>A</sup> $\pm$ 0.15
	B	0.33 <sup>A</sup> $\pm$ 0.02	189.45 <sup>B</sup> $\pm$ 0.51	2.90 <sup>A</sup> $\pm$ 0.01	10.83 <sup>B</sup> $\pm$ 0.02
	C	0.30 <sup>B</sup> $\pm$ 0.00	161.25 <sup>B</sup> $\pm$ 1.52	2.99 <sup>A</sup> $\pm$ 0.02	10.80 <sup>C</sup> $\pm$ 0.12
	D	0.28 <sup>C</sup> $\pm$ 0.01	141.05 <sup>B</sup> $\pm$ 0.55	3.01 <sup>A</sup> $\pm$ 0.10	10.63 <sup>A</sup> $\pm$ 0.17
Cashew	A	0.20 <sup>A</sup> $\pm$ 0.02	40.21 <sup>A</sup> $\pm$ 0.10	3.18 <sup>A</sup> $\pm$ 0.01	10.64 <sup>A</sup> $\pm$ 0.01
	B	0.23 <sup>B</sup> $\pm$ 0.01	36.11 <sup>A</sup> $\pm$ 0.11	3.16 <sup>A</sup> $\pm$ 0.01	10.55 <sup>A</sup> $\pm$ 0.01
	C	0.21 <sup>C</sup> $\pm$ 0.02	38.31 <sup>A</sup> $\pm$ 0.21	3.12 <sup>A</sup> $\pm$ 0.01	11.66 <sup>B</sup> $\pm$ 0.01
	D	0.24 <sup>C</sup> $\pm$ 0.01	38.42 <sup>A</sup> $\pm$ 0.32	3.20 <sup>A</sup> $\pm$ 0.01	11.66 <sup>A</sup> $\pm$ 0.01
Guava	A	0.23 <sup>D</sup> $\pm$ 0.01	21.85 <sup>B</sup> $\pm$ 0.03	3.36 <sup>A</sup> $\pm$ 0.02	12.37 <sup>C</sup> $\pm$ 0.03
	B	0.32 <sup>A</sup> $\pm$ 0.02	22.65 <sup>A</sup> $\pm$ 0.01	3.48 <sup>A</sup> $\pm$ 0.02	12.66 <sup>C</sup> $\pm$ 0.15
	C	0.25 <sup>A</sup> $\pm$ 0.01	20.93 <sup>A</sup> $\pm$ 0.06	3.28 <sup>A</sup> $\pm$ 0.08	11.87 <sup>A</sup> <sup>C</sup> $\pm$ 0.25
	D	0.21 <sup>C</sup> $\pm$ 0.02	22.85 <sup>C</sup> $\pm$ 0.00	3.26 <sup>A</sup> $\pm$ 0.01	12.56 <sup>B</sup> $\pm$ 0.25

*To be continued*



Continuation

Nectar	Mark	Titratable acidity ATT (g citric acid / mL)	Vitamin C (mg ascorbic acid/100 mL <sup>-1</sup> )	hydrogen potential pH	Soluble Solids (°Brix)
Mango	A	0.35 <sup>A</sup> ± 0.01	21.29 <sup>C</sup> ±0.60	3.35 <sup>A</sup> ± 0.01	12.36 <sup>A</sup> ± 0.00
	B	0.33 <sup>A</sup> ± 0.02	19.99 <sup>C</sup> ±0.50	3.58 <sup>A</sup> ± 0.01	12.56 <sup>A</sup> ± 0.00
	C	0.28 <sup>B</sup> ± 0.01	18.17 <sup>C</sup> ±0.40	3.53 <sup>A</sup> ± 0.00	12.93 <sup>AB</sup> ± 0.06
	D	0.32 <sup>C</sup> ± 0.02	22.15 <sup>A</sup> ±0.60	3.37 <sup>A</sup> ± 0.01	12.49 <sup>B</sup> ± 0.20
Passion fruit	A	0.56 <sup>A</sup> ± 0.02	18.2 <sup>A</sup> ±0.00	3.32 <sup>A</sup> ± 0.01	11.34 <sup>A</sup> ± 0.11
	B	0.46 <sup>B</sup> ± 0.01	17.92 <sup>A</sup> ±0.50	3.03 <sup>A</sup> ± 0.02	11.56 <sup>B</sup> ± 0.10
	C	0.45 <sup>C</sup> ± 0.01	22.05 <sup>B</sup> ±0.78	2.98 <sup>A</sup> ± 0.01	12.58 <sup>A</sup> ± 0.10
	D	0.55 <sup>A</sup> ± 0.01	20.15 <sup>B</sup> ±0.48	3.11 <sup>A</sup> ± 0.00	12.45 <sup>C</sup> ± 0.20

Averages followed by the same letter in the column do not differ statistically at 5% probability by the Tukey test.

Table 01. Analyzes of titratable acidity, Vitamin C, pH and ° Brix in nectars of brands A, B, C and D

Normative Instruction N °. 12, of September 4, 2003				
Néctar	Titratable acidity ATT (g citric acid / mL)	Vitamin C (mg ascorbic acid/100 mL <sup>-1</sup> )	hydrogen potential pH	Soluble Solids (°Brix)
Acerola	Mínimo de 0.20	Mínimo de 160.00	-----	Mínimo de 10.00
Cashew	Mínimo de 0.12	Mínimo de 15.00	-----	Mínimo de 10.00
Guava	Mínimo de 0.10	Mínimo de 14.00	-----	Mínimo de 10.00
Mango	Mínimo de 0.20	-----	-----	Mínimo de 10.00
Passion fruit	Mínimo de 0.25	-----	-----	Mínimo de 11.00

Table 02. Normative Instruction N°. 12/2003

For the industrial processing of nectar, the high content of titratable acidity is important, as it reduces the need for adding acidifiers and provides food security, making it difficult to develop yeasts (LIMA et al., 2002). Normative Instruction 12, of September 4, 2003, does not establish values for the vitamin C content in passion fruit and mango nectars. However, due to the benefits brought to the human body by vitamin C, this parameter was performed in order to verify the amount present in the passion fruit and mango nectar.

All samples, except acerola, had a vitamin C content below the recommended daily requirement for adults and currently recommended is at 45/mg according to RDC No. 269 of 2005 from the National Health Surveillance Agency (BRASIL, 2005).

Vitamin C degrades easily, and the factors that most affect its stability are the alkaline environment, heat, light, metals (Fe, Cu and Zn) and ascorbate oxidase enzyme (FARAONI et al., 2010; CHIM; ZAMBAZI; RODRIGUES, 2013).



Although the pH is not regulated by the Brazilian legislation for fruit nectars, its determination in drinks is extremely important, since it is an parameter of easy and quick evaluation. It should never be higher than 4.5, since above this value it can favor the growth of *Clostridium Botulinum* (DAMIANI et al., 2011; MIRANDA et al., 2015). Some factors such as the time of pasteurization and the action of microorganisms can affect the pH of nectars (CHIM; ZAMBIAZI; RODRIGUES, 2013).

The pH values can directly affect the color of the product and consequently its acceptance by the consumer (SILVA et al, 2016).

The results obtained by microbiological analysis showed values in accordance with Consolidation Ordinance No. 5 of September 28, 2017. The legislation establishes the absence of total and thermotolerant coliforms regardless of the number of samples analyzed.

## CONCLUSIONS

The acerola, cashew and guava nectars analyzed presented the parameters total titratable acidity, soluble solids and Vitamin C within the quality standards established by Normative Instruction No. 12, of September 4, 2003 MAPA. The potential of hydrogen (pH) and vitamin C for mango and passion fruit nectars is not established by legislation.



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# EVALUATION OF PHYSICAL-CHEMICAL AND MICROBIOLOGICAL PARAMETERS OF WATER IN SHALLOW WELLS LOCATED IN NEIGHBORHOODS OF PARADISE TOCANTINS

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## ABSTRACT

The work evaluated and compared the physical-chemical and microbiological quality of water from residential shallow wells in 4 (four) neighborhoods located in the municipality of Paraíso do Tocantins - TO with the treated water consumed by the population. The physico-chemical analyzes of color, hydrogen potential (pH) and turbidity followed the analytical methods of the AWWA (America Water Works Association) Standart Methods for Examination of Water and Wastewater and for the microbiological procedures for determination of total and thermotolerant coliforms were used the multiple tube technique, as described by the National Health Foundation - FUNASA. The results of the analyzes were compared with the standards established by Consolidation Ordinance No. 5, of September 28, 2017. To check if there was a significant difference between the results, ANOVA and the Tukey test

were applied at the level of 5% of significance. The statistical analysis identified a difference between the turbidity of the analyzed wells. The results obtained showed that the microbiological analyzes of the water from the researched wells showed a level of bacterial contamination, making them unfit for human consumption, since the values found were in disagreement with Consolidated Ordinance No. 5, of September 28, 2017, from the Ministry of Health, which sets the standard for drinking water for human consumption.

### **Keywords:**

*Coliforms, environmental impact and public health*



# INTRODUCTION



The wells most used to collect groundwater are shallow wells, which are commonly drilled by hand and given different names throughout Brazil, such as the Amazon well, cistern, cacimba, cacimbão, caipira well or simply “well” (SILVA et al, 2014; MOTTA JG, et al. 2014). They have been an alternative for people to consume water from underground reserves (MOTTA J.G, et al. 2014; KUHN M; ZART; OLIVEIRA, 2015).

Groundwater, is generally, less contaminated by biological and chemical factors than surface water sources, as they are not exposed to various polluting agents (KUHN; ZART; OLIVEIRA, 2015). Water is one of the most important natural resources, essential to human life and activities due to its functions in public, industrial, agricultural and preservation of aquatic life and can serve as a vehicle for various biological and chemical agents (KUHN; ZART; OLIVEIRA, 2015). The water with crystalline appearance and without odor may not be free from harmful microorganisms (ROCHA E.S, 2010; YAMAGUCHI, 2013). The microbiological analysis of water identifies microorganisms harmful to human health, which are commonly found in the intestinal tract of warm-blooded animals (ROCHA E.S, 2010).

*Escherichia coli* is a pathogenic microorganism of fecal origin devoid of free life in the environment, which when present in water indicates contamination by feces (YAMAGUCHI, 2013). To detect whether the water is within the standards for consumption by the population, physical-chemical and microbiological analyzes established by Consolidation Ordinance No. 5, of September 28, 2017, of the Ministry of Health (BRASIL, 2017) are used. Monitoring water quality through physical, chemical, bacteriological and organoleptic analyzes helps to detect water pollution and contamination problems, helping treatment and minimizing the use of chemical substances for their purification (STRUCKMEIER et al; 2005).

The objective of this study was to compare the physical-chemical and microbiological parameters of water from residential shallow wells in 4 (four) neighborhoods located in the municipality of Paraíso do Tocantins - TO and the treated water consumed by the population and compare them with the Consolidation Ordinance No 5, of September 28, 2017 (BRASIL, 2017).

## Material and methods

The collections took place between March and December 2018, in the morning between 7 am and 9 am, in the Pouso Alegre, Jardim Paulista, Milena and Setor Oeste neighborhoods, in the city of Paraíso do Tocantins. In each neighborhood, one (1) residence with an artesian well was drawn, totaling four (4) monthly collections for eight (8) months. Figure 01 shows the locations for collecting water samples in shallow residential wells.



Figure 01. Groundwater abstraction from shallow wells in Paraíso do Tocantins  
Source: Google Earth, 2018.

The procedures adopted for the collection and transportation of the samples followed the procedures described by the National Health Foundation (FUNASA, 2006). 1000 ml of water were collected in sterile polyethylene flasks for physico-chemical analysis and 100 ml in sterile glass flasks containing 1 ml of 0.1 N sodium thiosulfate solution for microbiological analysis. The samples were placed in a thermal box with a temperature equal to or less than 8 °C and transported to the Analytical Chemistry Laboratory of the Federal Institute of Education, Sciences and Technology of Tocantins - Campus Paraíso do Tocantins for analysis of the samples. The analyzes of color, hydrogen potential (pH) and (pH) turbidity followed the analytical methods of the AWWA (America Water Works Association) Standart Methods for Examination of Water and Wastewater (APHA, 2005) and analyzes of total and thermotolerant coliforms were performed using the multiple tube method, according to procedures described by the National Health Foundation and confrontation with the rules established by Consolidated Ordinance No. 5, of September 28, 2017 (BRASIL, 2017).

The physical-chemical and microbiological analyzes were processed in triplicate and their results were submitted to analysis of variance. To check if there was a significant difference between the results, ANOVA was applied and between the means of the response variables, the Tukey test at the level of 5% of significance. All statistical analyzes were performed using the Sisvar version 5.6 program (FERREIRA, 2019).

## Results and discussion

Table 01 presents the results of the parameters analyzed as established by Consolidation Ordinance No. 5, of September 28, 2017.



Physical parameters chemical and microbiological	shallow well A	shallow well B	shallow well C	shallow well D	potable water	Ordinance n°5/2017MS
Color	2,38 <sup>A</sup> ± 1,51	2,19 <sup>A</sup> ± 0,88	2,25 <sup>A</sup> ± 1,34	1,89 <sup>A</sup> ± 1,34	2,48 <sup>A</sup> ± 1,66	15 uH
pH*	6,91 <sup>A</sup> ± 0,09	6,93 <sup>A</sup> ± 0,14	6,91 <sup>A</sup> ± 0,12	6,87 <sup>A</sup> ± 0,09	6,86 <sup>A</sup> ± 0,12	6,0 e 9,5
Turbidity	0,64 <sup>A</sup> ± 0,16	0,58 <sup>A</sup> ± 0,17	0,36 <sup>B</sup> ± 0,10	0,32 <sup>B</sup> ± 0,15	0,24 <sup>B</sup> ± 0,08	5,0 NTU
CT** (NMP/mL)	22,88 <sup>A</sup> ± 4,52	21,25 <sup>A</sup> ± 3,69	19,50 <sup>A</sup> ± 4,00	22,00 <sup>A</sup> ± 4,28	0,00 <sup>B</sup> ± 0,00	absence
CTT*** (NMP/mL)	34,13 <sup>A</sup> ± 7,41	30,25 <sup>A</sup> ± 12,38	31,75 <sup>A</sup> ± 7,83	29,50 <sup>A</sup> ± 7,31	0,00 <sup>B</sup> ± 0,00	absence

Means followed by the same letter in the lines do not differ ( $p \geq 0.05$ ). pH \* Hydrogenionic potential. CT \*\* Total coliforms. CTT \*\*\* Thermotolerant coliforms.

Table 01. Results of physical, chemical and microbiological parameters of water from shallow wells analyzed.

According to Table 01, the results of the physical chemical analyzes of the water samples from wells A, B, C and D were in accordance with the values established by Consolidation Ordinance No. 5, of September 28, 2017, of the Ministry of Health. Microbiological analyzes for total coliforms and thermotolerant coliforms showed disagreement with the above-mentioned legislation and with treated water. One of the possible causes of contamination of the waters of the researched wells is due to the fact that they do not have external protection (cover) and are located close to the septic tanks. The presence of coliforms in the analyzed waters may be linked to the poor construction conditions of the wells and the proximity of septic tanks that can proliferate pollution by fecal material of human origin (GONÇALVES, et al., 2005). In studies by Silva et al (2003), in Feira de Santana-BA, in shallow wells, manually drilled, the authors found high percentage values for samples with the presence of coliforms. Results similar to that of groundwater contamination research in shallow wells by total and thermotolerant coliforms were observed by Motta et al. (2015) that in the Middle Valley Region of Itajaí - SC, reported the presence of total coliforms in 25.53% of the analyzed samples, being 8.51% determined as Thermotolerant Coliforms. Similar data are also reported by Costa et al. (2012), who, when assessing the quality of groundwater in wells in the state of Ceará, found 40% presence of Total Coliforms and 12.2% presence of *Escherichia coli* in the analyzed samples. Costa et al. (2014), when assessing the water quality of the underground water source in urban areas of Vitória da Conquista, BA found values of pH, turbidity and thermotolerant coliforms within the limits of the legislation.

Oliveira et al. (2018), performing physical-chemical and microbiological analysis of water from artesian wells for independent use, found results for the physical chemical parameters color and turbidity within the stipulated by the legislation and for the values for the hydrogen potential pH in disagreement with the legislation. Regarding the microbiological result of Oliveira et al. (2018), found 70% of the samples were collected without disagreement with the recommendations of Consolidation Ordinance No. 5, of September 28, 2017. Kuhn, Zart and Oliveira (2015) assessing the quality of the water in the artesian wells that supply the District of Boa Vista, in the Municipality of Triunfo - RS, find variations in pH showing a tendency from neutral to alkaline, they were unable to detect the apparent color, average turbidity equal to 5.25 NTU and 40% contamination by total





coliform coliforms and *Escherichia coli* in the collected water samples. Zamilian, Paula and Zamilian (2018), conducting a Microbiological evaluation of Artesian Well Waters in Rural Properties in the Municipality of Colorado do Oeste - Rondônia, found 30% of the samples contaminated with Total Coliforms as well for Thermotolerant Coliforms.

Evaluating the results obtained, the presence of total and thermotolerant coliforms was verified in all water samples collected in wells A, B, C and D. According to what is established in Consolidation Ordinance No. 5, of September 28, 2017 (BRASIL, 2017), if any of these microorganisms are present, water cannot be consumed without previous treatment, to be carried out with some antibacterial agent, such as, for example, chlorine (KUHN; ZART; OLIVEIRA, 2015).

## CONCLUSION

The results obtained showed that the physical and chemical analyzes were in compliance with the legislation, but all water samples from the wells surveyed showed bacterial contamination making them unfit for human consumption. Consolidation Ordinance No. 5, of September 28, 2017, determines that water should not be consumed without previous treatment with antibacterial agent when total or thermotolerant coliforms are present. Thus, it is necessary to implement measures to manage the use of groundwater, monitor and treat the quality of water in these neighborhoods, and carry out actions on environmental education to eradicate the sources of contamination and the spread of diseases caused by the consumption of this water.



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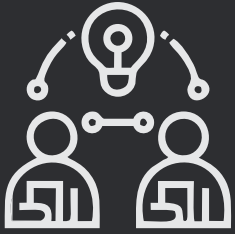
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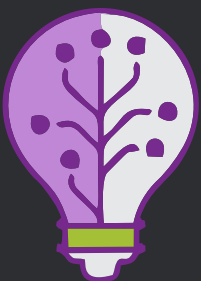


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